DRUM *and* CROAKER

A Highly Irregular Journal for the Public Aquarist

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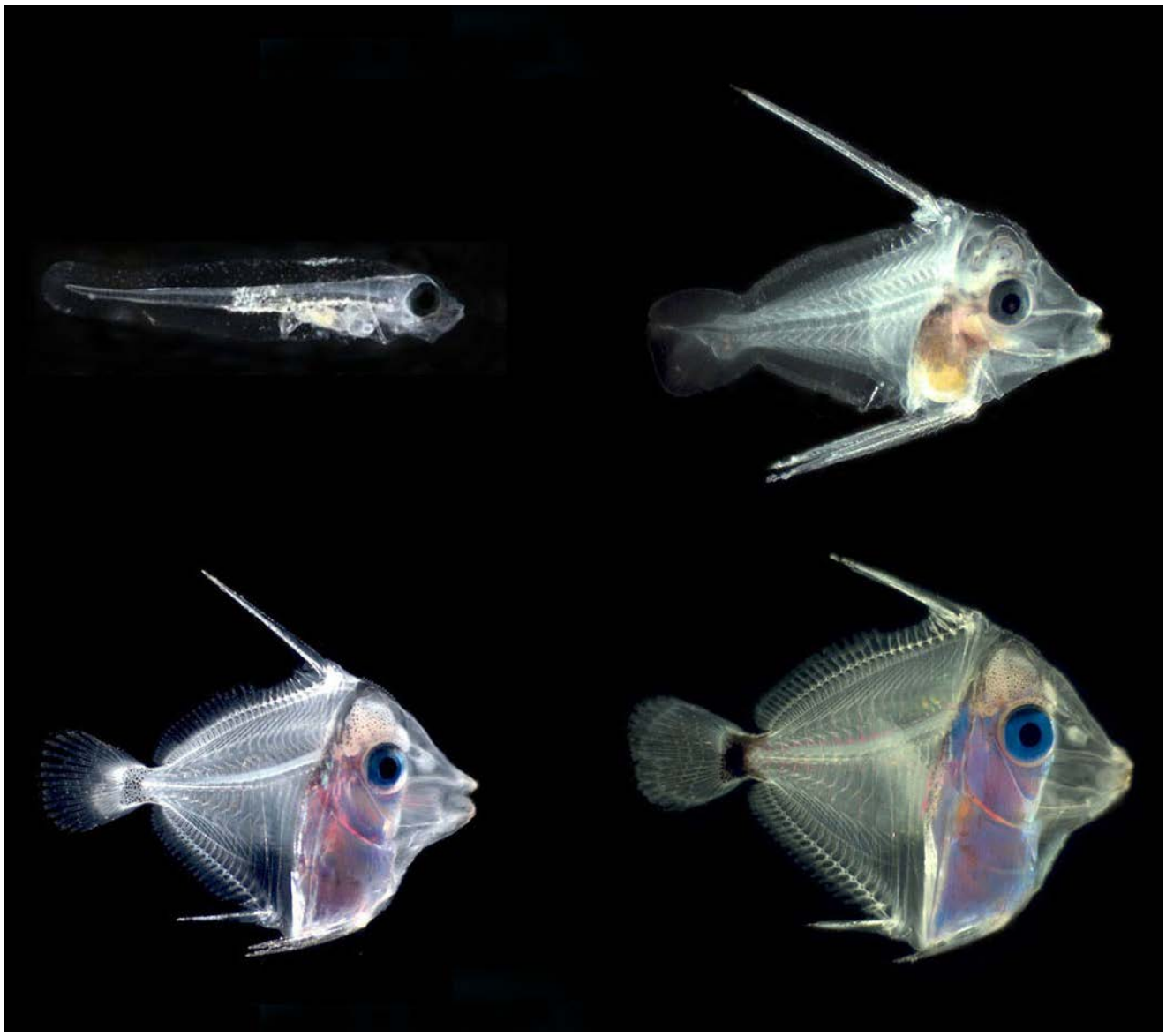

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DRUM AND CROAKER ~50 YEARS AGO

Excerpts from Volume Sixty-Six (LXVI) 1966,
Edited by John H. Prescott, Curator, Marineland of the Pacific
Richard M. Segedi

UNDERSEA GARDENS IN SEATTLE OPENED TO THE GENERAL PUBLIC

Eric Friese, Curator, Undersea Gardens, Seattle

Basically the Undersea Gardens consists of a modified ship's hull with 112 large viewing windows below the water line. Large alcoves with windows on each side protrude from the hull into the display area. From the outside of the hull a 15 foot steel ledge (about 7 feet below the surface) extends outward around the entire hull, and ends in an 8 foot high plexiglass panel fence. For easy maintenance (i.e. the removal of algae, barnacles, etc.) these 1/4 inch panels slide in and out in small grooves on each side of the steel braces. A small gap in the center, running lengthwise, permits the removal of the plexiglass panels.

An important factor in the uniqueness of this exhibit is the tremendous panoramic view afforded the visitor. Rather than looking into individual small tanks, as presented in the conventional public aquarium, the visitor is completely surrounded by one continuous enclosure, featuring an entire cross section through the various habitats of marine life found in Puget Sound. With such a gigantic display area to fill, heavy emphasis is placed on large, natural rock reefs, kelp beds and well designed fiberglass octopus caves, which are camouflaged to blend in perfectly with the surroundings.

MAINTENANCE PROBLEMS IN LARGE PUBLIC AQUARIA

Frank de Graaf, Curator, Aquarium and Reptile House, Natura Artis Magistra

Beside the accumulation of inorganic salts another chemical constituent of the seawater is thought to be an important agent in the so-called "aging" process of the circulating seawater. This chemical constituent is not known, but is believed to be some organic compound which is not broken down and which is further responsible for the yellow-brown colour of "aged" seawater. By removing this colouring from the water, this water is again made suitable for sustaining aquatic life. To minimize the chemical changes in the aquaria brought about by the activity of the animals, the rate of flow of the water through the aquaria has to be as large as can be realised with the existing installation. Additional aeration is necessary in the large seawater tanks.

ANTHIUM DIOXIDE - A REVELATION

Warren Zeiller, Curator of Fishes, Miami Seaquarium

A revolutionary water additive soon will be available under the trade name Krystal Klear. This liquid chemical compound is an Anthium Dioxide complex, the basis of which is chlorine. Chlorine in its many forms is fine in swimming pools and drinking water, but always has proved a deadly addition for all but mammalia in aquatic environments. The new product is no more difficult to maintain than those for copper compounds, harmless and even beneficial to aquatic vertebrata. Unfortunately, like copper compounds, it is not tolerated by most invertebrates.

ON THE ART OF KURAGEDO: A PHILOSOPHY FOR JELLY TEAMS

Chad Widmer PhD, chad.widmer@pdza.org

Point Defiance Zoo and Aquarium, 5400 Pearl Street, Tacoma, WA, USA

The Point Defiance Zoo and Aquarium did not have a jellyfish gallery when I joined the team. Instead we had one little moon jelly exhibition that wasn't particularly loved or well cared for. It was often the lowest ranking aquarist who was relegated to caring for them. But things have changed! Last year we had the pleasure of acquiring funding for, designing, and building a brand new little jellyfish exhibition. What I like most about this gallery is that at this moment it is possibly one of the most flexible jellyfish exhibitions in the world. We can display any type of jellyfish body plan, originating from anywhere in the world, from the deep sea to the tropics. The gallery has a dance floor and a rocking wi-fi enabled sound system. It also has networked signage that we can modify from our phones. It's fair to say we're proud of it, especially since during 2015 PDZA enjoyed its highest recorded attendance in its venerable 100 year history. But more importantly, I am very proud of the Defiant Jellies Team, and how we have come together to become fully functional and self-reliant in such a short period.

As you may be acutely aware, maintaining a jellyfish gallery very much requires a sustained and prolonged team effort. A solitary jelly keeper left to tend the gallery by themselves will burn out, and move on in two years. To avoid this sequence of events myself I set about the process of training a new team of jelly keepers who are 'with it' and 'for it.' When I train a new jelly-head I require that they master a skill before they are allowed to learn the next one. Some of you reading this may have experienced 'the Widmer way.' I hope you found it time well spent. This method ensures that everyone on the team knows how to do everything to a high standard, and if someone has told me they've done something, I know that they have done it well without needing to double check. The method also allows me to confidently take weekends and vacations without returning to disaster. It also allowed me to go off to graduate school in Scotland without having the jelly galleries at MBA crash behind me when I left.

During the new exhibition build out process at PDZA the Team learned each new jellyfish husbandry skill with quickness, and they were eager to progress to the next level. I began to ask myself, what is it about this particular team that makes everyone so interested in learning so rapidly? And why are they so good at it? And why is doing everything to a very high standard really important to these particular individuals? I should also say that they all self-selected and chose to join the jelly team, they weren't mandated to. In our service area conversations I soon learned that all of us had some type of martial arts background in common. Other international jelly keepers I know turn out to be martial artists as well. We all value quality, discipline, and paying attention to detail. Martial artists also understand and value mastering a skill before being allowed to progress to the next one, and these traits serve jelly keepers well.

It was from these conversations that the art of Kuragedo was born. 'Kurage' is the Japanese word for jellyfish, and 'do' means* 'the way.' Kuragedo is the way of learning to care for jellyfish. It is the art of training a new jelly keeper, and it is a method for knowing the

relative skill set of a potential new hire. It is also the art of constructing and maintaining all things jellyfish gallery and culture related. Teamwork, mutual respect, and trust are the foundations on which Kuragedo are built.

Similar to most martial arts, a code of conduct and ethics exists amongst the practitioners of Kuragedo. We call them the tenets. It is not a static list, but naturally a fluid one. We are open to suggestions and contributions as The Art grows.

The tenets of Kuragedo are:

1. To be better aquarists today than we were yesterday.
2. Recognize that jelly keeping is a process, not an event.
3. We value trust and brotherhood among Teammates.
4. We aim to display the most diverse, healthiest, and best looking collection in the world.
5. We will not be products of our environment. Our environment will be a product of us.
6. Take the initiative, and make it happen.
7. It doesn't matter where you come from. It matters where you are going.
8. A mind is a terrible thing to waste.
9. We celebrate our mistakes, and learn from them.
10. *Semper laborantes!*

Kuragedo is about establishing and maintaining a positive, fun and productive team culture that can accomplish any goal it sets for itself. A white belt in Kuragedo can walk around the gallery without accidentally destroying anything. A yellow belt can harvest *Artemia* nauplii and rotifers, and feed them to the appropriate animals. A green belt knows the scientific names of things, and can bleach a kreisel whilst a black belt can start new cultures and sustainably keep a gallery going. A master dreams up new things that make us all reach for our cameras. As for the color of my belt… all of the black has worn off, leaving me a white belt once again. My Team and I are evolving.

**Some words of acknowledgement about the word 'Do.' In the Japanese language, the word 'do' has far more profound meanings than simply, 'the way.' Your homework Teammate is to find out how and report back.*

RISING TIDE CONSERVATION – 2016 REVIEW

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Rising Tide Conservation promotes reef conservation through advancing marine ornamental aquaculture. In the short time it has been around, it has grown from a cool idea to a driving force. Established in 2009, the program is based on a diverse base of stakeholders. The heart of the program is the research team. These folks are supported by display aquaria, and hobbyist industry members including retailers and wholesalers, as well as committed hobbyists, themselves. Focusing research at facilities with time, space, and personnel dedicated to aquaculture allows fast track developments. Communicating those developments is the key to making them sustainable. Sustainability is when marine ornamental fish techniques are incorporated into commercial production by anyone with the dedication to follow the trailblazers.

So what's happening and where? Florida is home to three separate facilities – the Tropical Aquaculture Lab of the University of Florida in Ruskin, The agricultural extension laboratories of the University of Florida in Vero Beach, and the Florida Keys Community College in Key West Florida.

The Tropical Aquaculture Lab (TAL) has a variety of folks all working towards the same goals. The programs at TAL are all under the direction of Matt DiMaggio. Matt trains students and directs larval trials. Current broodstock being held at the TAL include, Pacific Blue Tang (*Paracanthurus hepatus*), Yellow tang (*Zebrasoma flavescens*), Milletseed Butterflyfish (*Chaetodon miliaris*), Bartlett's anthias (*Pseudanthias bartlettorum*), Emperor angelfish (*Pomacanthus imperator*), Semicircle angelfish (*Pomacanthus semicirculatus*), Melanurus wrasse (*Halichoeres melanurus*), Yellow wrasse (*Halichoeres chrysus*),and two species of schooling bannerfish (*Heniochus diphreutes, Heniochus acuminate*). Larval rearing trials are being continuously conducted dependent upon egg production from the species. In 2016, larval culture efforts are aimed at adapting rearing systems that have been used for the successful culture of yellow tangs to Pacific Blue Tang larval culture protocols. They worked! In the summer of 2016, just two weeks after the opening of the iconic movie "Finding Dory" Eric Cassiano and Kevin Barden of the tropical aquaculture lab in Ruskin announced that they had successfully broken through the bottlenecks in rearing this species.

Eric Cassiano's focus on increasing the production (and availability) of the copepod *Parvocalanus crassirostris* using new production systems and management techniques was a key element in the success. This key, when paired with Kevin Bardens's focus on broodstock maintenance and larval rearing is how we get from success to repeatability. The larval development timing had slight variations, but in general involved newly hatched Pacific Blue Tangs measuring just under two millimeters long, with no eyes or mouth drifting for two days while they absorbed their yolk. During that time they develop eyes and a mouth. Survival rates

to day 4 were as high as 80% or better. The gape at day 4 is 40-50 microns. That's smaller than a period on this page. The team produced massive amounts of copepods and other live food, managed water quality and character, feeding, and lighting. As the days and weeks ticked by, the fish started behaving and growing like nothing seen before. At day 40 post hatch, the fish had started to settle and looked like small, colorless replicates of their parents. On day 51 the first baby "Dory" was photographed, not on an Indo Pacific reef, but in a greenhouse in Ruskin, Florida (figure 1).

Down in Vero Beach, Cortney Ohs has taken his work in bait fish propagation and applied that learning to ornamentals - both with in-house broodstock and with larval rearing from eggs collected at display aquaria. The folks in Vero Beach have broodstock of blue chromis (*Chromis cyanea*) and Pacific blue tangs (*Paracanthurus hepatus*). This facility is the home for eggs collected at display aquariums and shipped for larval culture. Because of this, on any given day they may have 5-10 different species growing in numbers from 10 to 100 to 1000 larvae.

In the Florida Keys is the newest member of the Rising Tide Conservation research collaborative. Dr. Patrick Rice and Dr. Michelle "Mick" Walsh are not only training the next generation of marine aquarists, they are teaching them the skills needed for marine aquaculture. Their program began as many do with a focus on clownfish and dotty backs. But in 2015, they explored the potential for several new Caribbean species including the four-eyed butterflyfish (*Chaetodon capistratus*) and the rock beauty angelfish (*Holacanthus tricolor*). Now, in 2016, with the help of Rising Tide Conservation, the FKCC team has developed student internships to focus on the previously mentioned Caribbean species, while also expanding student research projects to the Indo-Pacific Yellow Coris Wrasse (*Halichoeres chrysus*), development of Caribbean coral production system, and the development of an innovative mesocosm zooplankton production system for marine ornamental larval rearing.

Over in Hawaii, the team at the Oceanic Institute (OI) of Hawaii Pacific University made the biggest splash of 2015. That team is led by Dr. Chad Callan. Chad and his team worked for many years on cracking the challenges of yellow tang rearing. In late 2015, they struck gold. Three broods of eggs from broodstock were successfully brought through all stage of larval development to juveniles. The first brood had less than 100 make it through but the second had around 700 – a pretty good improvement. Work on replicating and documenting this success continued in 2016 along with hosting a team of Asian producers to complete the process of advancement to commercial production for the Pacific Blue Tang.

But the Oceanic Institute was not to be outdone by the University of Florida in 2016. Last year saw OI take on its first intern/fellow, Avier Montalvo. Avier worked with eggs collected from a large, local display aquarium tank. He used the management lessons from the yellow tang to move forward work on tangs, angels, and wrasses. In one year, his project saw the production of more yellow tangs, Klein's butterflyfish, Milletseed butterflyfish (figure 2), Potter's angelfish (figure 3), and the Hawaiian cleaner wrasse.

In 2017, Avier plans to switch to the research station in San Diego at the Hubbs-SeaWorld Research Institute (HSWRI) to continue novel larval rearing based on egg collections

from display aquaria. 2016 saw the entry of HSWRI into the Rising Tide program with the broodstock set up of two Centropyge angelfish species.

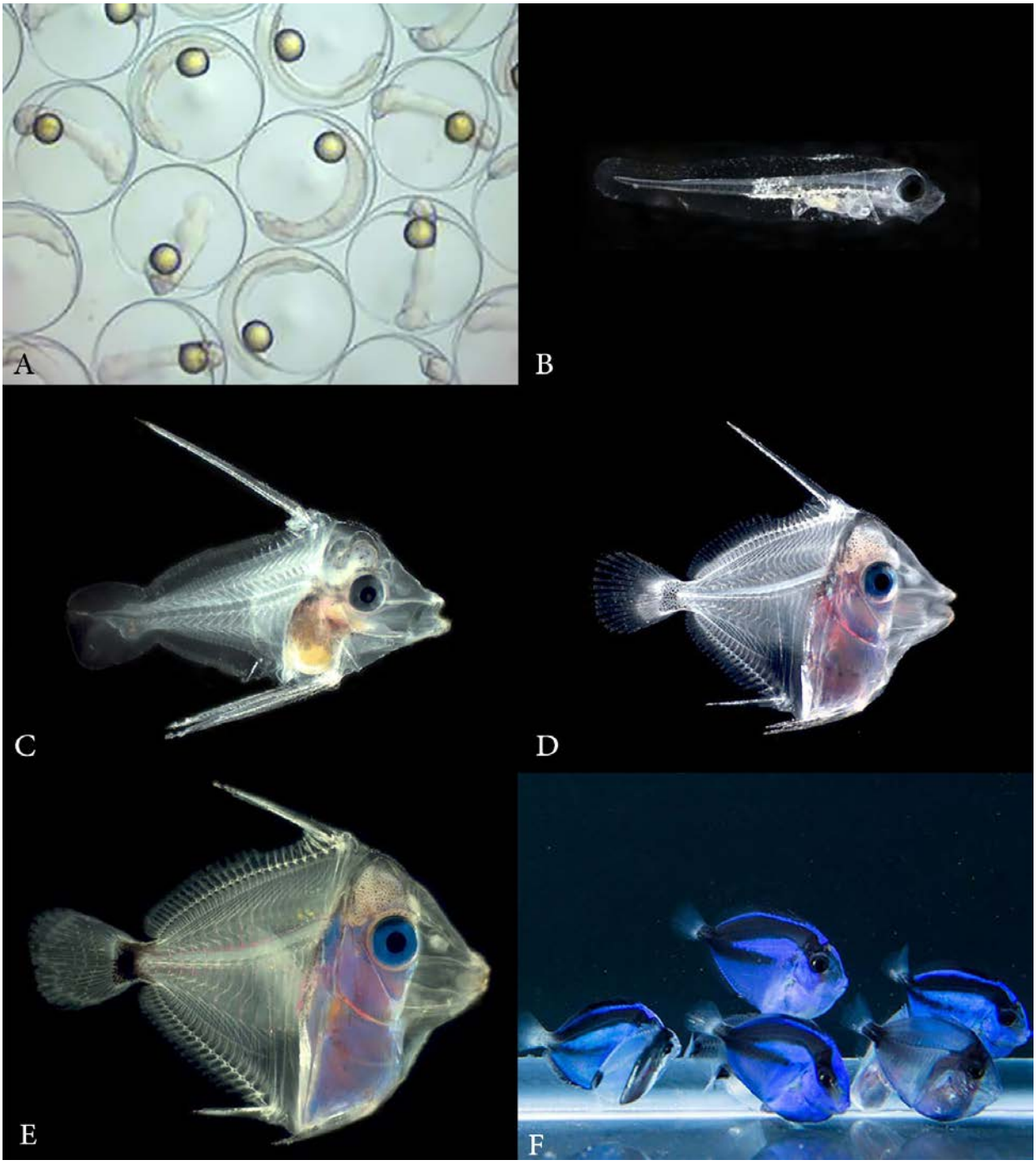

Figure 1. Larval Progression of the Pacific Blue Tang (*Paracanthurus hepatus)*. A.Day 5 PH, B. Day 19 PH, C. Day 29 PH, D. Day 33 PH, E. Day 55 PH

Rising Tide continues to evolve. The goal remains to go from rearing to implementation by commercial producers for sustainability. Sustainable production provides alternatives to reef collecting. The researchers for Rising Tide Conservation continue to focus on rearing techniques for difficult species. Beyond this, they share their successes at meetings and in popular and scientific publications. Increasing numbers of display aquaria across the US contribute to this work by telling about reef challenges in signage and tours, creating tanks dedicated to aquacultured fish, supplying collected eggs and financially supporting researchers and students. You can be part of the program. Learn more about Rising Tide on our Facebook page or contact me at judy.st.leger@seaworld.com to learn how you can be part of this cool project.

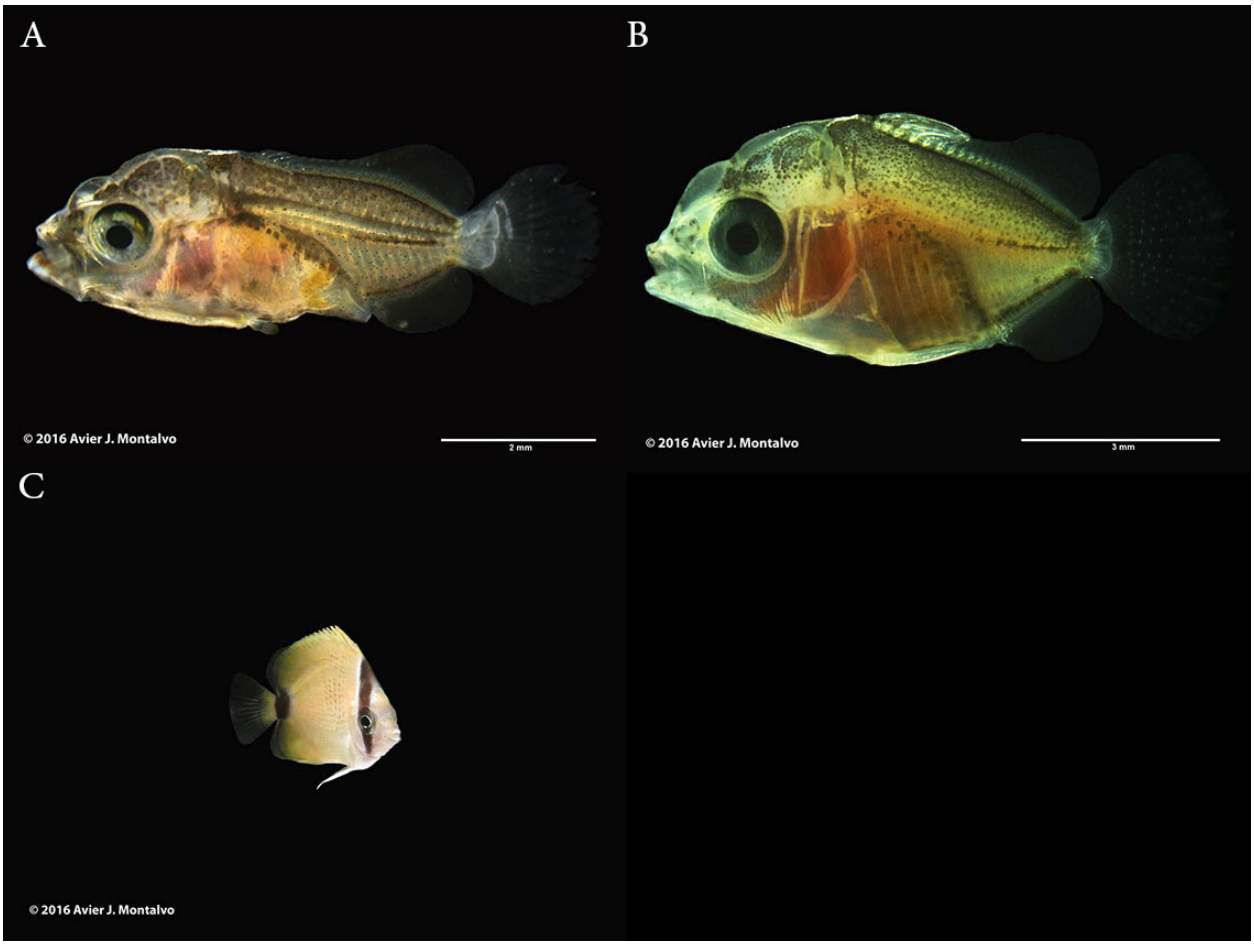

Figure 2. Larval progression of the Milletseed Butterflyfish (*Chaetodon miliaris*) a. 25 DPH, B. 40 DPH, C. 88 DPH

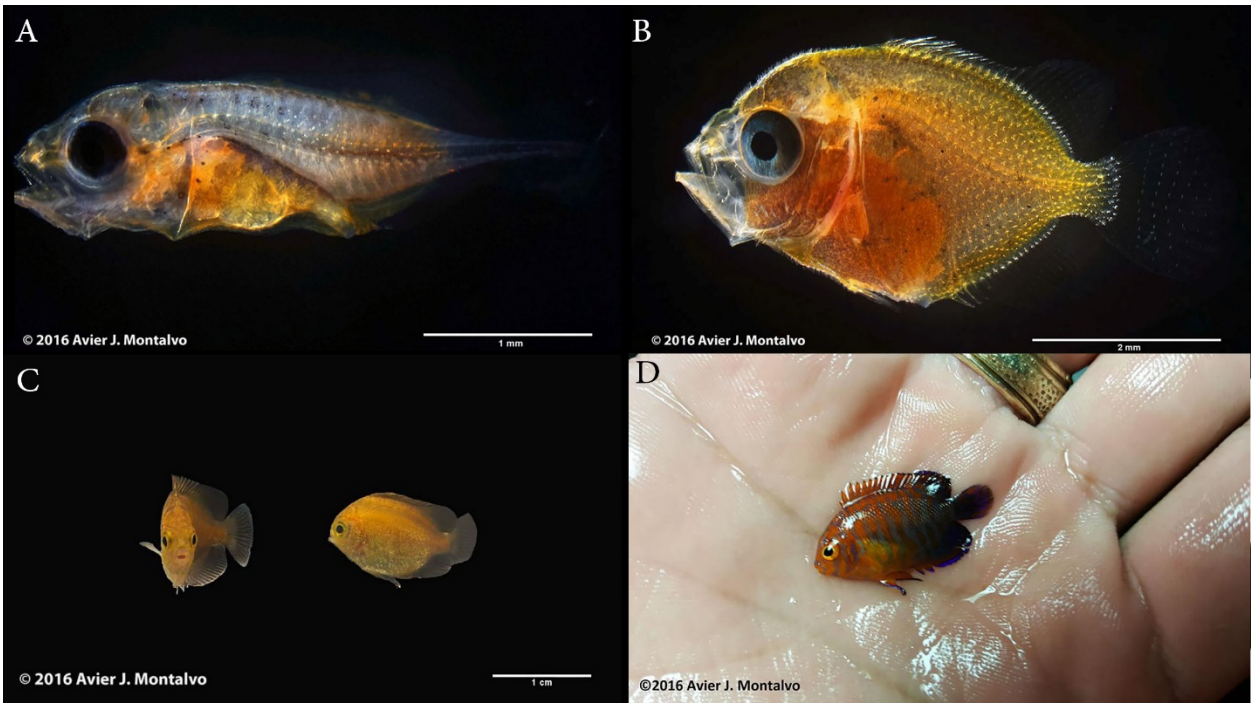

Figure 3. Larval progression of the Potter's Angelfish (*Centropyge potteri*). A. 10 DPH, B. 33 DPH, C. 59 DPH, D. 73 DPH

OBSERVATIONS ON THE SUBJECTIVITY OF EPAULETTE SHARKS *(Hemiscyllium ocellatum)* TO TEMPERATURE DEPENDENT SEX DETERMINATION

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Introduction

In 2005 the Tennessee Aquarium opened its saltwater building, Ocean Journey which is the perfect complement to River Journey, the larger freshwater themed building next door. This building includes a 9,000 gallon elasmobranch touch experience housed in the tropical greenhouse-like top floor. Many different species are found in this exhibit, but the focus of this article and this aquarist's favorite individual animals, are a pair of adult wild collected Epaulette Sharks *(Hemiscyllium ocellatum).*

Fast forward six years to Halloween, 2011; my first day as an aquarist at the Tennessee Aquarium. My introduction to my new exhibit included hand feeding the female Epaulette Shark that was now under my care. There were plenty of other elasmobranches vying for my attention, but she quickly won me over with her social and curious nature. She was the sole *ocellatum* on exhibit; the male shark was isolated in a backup system at the time.

Since both animals were being maintained similarly, I decided to add the male back to the main exhibit. After being apart (and having not bred previously) for over a year the duo wasted no time and began producing fertile eggs within a month. The female would deposit an average of four egg cases a month, two at a time, either on the substrate of the exhibit or amongst the loose rock piles. These eggs were removed from the exhibit and allowed to develop in a more controlled environment, and eventually baby sharks would emerge (Figures 1, 2, and 3…because this much cuteness deserves three pictures). It was noted, however, that moving newly deposited eggs to a tank with slightly different water parameters without a "stabilization period" would cause all development to cease and the yolk to rot, regardless of the care taken in transport and acclimation. I had initially assumed that the eggs would be more durable and less sensitive to moving to a new system earlier on in their development before slits began to form in the egg case. This was disproved by the loss of a group of eggs I moved within several days of being laid. It was therefore made general practice to keep the eggs in a small tank plumbed into the same system as the exhibit (Tank A) for a minimum of 20-30 days before moving them to another tank more suitable for the neonates to hatch out in. This stabilization period proved adequate and embryos continued development even after being moved to the hatching tank on a separate system.

After producing 35 pups, I was sufficiently enamored with the species, their reproduction, and the rearing of the pups. I decided to keep some animals to return to exhibit, as well as surplus some to other facilities. Up until this point the temperature at which the eggs had been developing was selected simply to be close to that of the exhibit water. The exhibit was maintained at 77°f, so initially the egg tank was kept at a "close enough" temperature of 78°f. At this temperature the gender of the first 35 shark pups was split 20 males to 15 females, 57% male, 43% female. Out of curiosity the egg tank was warmed to 80°f to determine the effect the

slightly warmer temperature would have on incubation time, and a group of nine eggs were added over the course of two months. This warmer temperature decreased incubation time as expected, but more intriguing was the effect it seemingly had on the gender of the pups; all but two of the group were female. I was excited by the prospect that these eggs may have had their gender determined by the temperature of their environment, but determining if that was the explanation would require a more thorough and controlled examination.

Methods

The adult sharks were allowed to remain on exhibit and were fed and maintained the same as they had been previously. Breeding patterns remained consistent, as well as egg production. In order to allow the eggs to stabilize they were placed in the aforementioned back up tank (Tank A) for (30) days before being moved into the temperature varied tanks. Tank A contained a rack made from lighting egg crate and seaweed feeding clips in order to maintain differentiation between eggs (Figure 4). Each clip is assigned a number, and each egg's date recorded. After (30) days the eggs were transferred to either the warm or the cool water holding tank, (Tanks B and C, respectively) alternating every other egg. The target would be n=30 eggs.

Since the baseline figures were found using 78°f, Tank B was set three degrees warmer at 81°f, while Tank C was held three degrees cooler, 75°f. Both systems maintained similar water chemistry leaving temperature the only variable. Identical egg tanks were plumbed into each system, consisting of a 10 gallon acrylic aquarium with an egg rack placed in the bottom consisting of seaweed feeding clips held to a PVC sheet with nylon nuts and bolts (Figure 5). This system allowed the eggs to be individualized and monitored, while still allowing them to hatch naturally. When a pup was found in an egg tank, it was removed, sexed, and added to an adjoining tank containing other pups. The eggs were then examined until the empty one was found, its date and sex data recorded.

Results

From 3/11/2014 until 10/5/2014, (30) eggs were removed from the exhibit and handled in this manner, producing 26 pups. The results can be seen in the table below. As a matter of curiosity, the effect the water temperature has on incubation times was also monitored, and is included in the table as well.

	Male	Female	Mean Incubation
Tank B (81°f)	8 (66.67%)	6 (33.33%)	118.9 days
Tank C (75°f)	9 (75%)	3 (25%)	169.1 days

these initial trials provided similar male biased results to our earlier 78°f incubation tests which led me to develop a new theory; perhaps the sex of the shark is determined within the first 30 days after the egg is deposited. It would therefore be influenced by the temperature at which Tank A is being maintained during the egg's initial stabilization period.

Methods, 2.0

The tests were run again; all variables were kept identical, with only the stabilization period being altered. The eggs were maintained in Tank A for only (14) days prior to being moved to their assigned temperature tanks. The target would be n=30 eggs, as before.

Results, 2.0

Eggs for this group were collected from 10/15/2014 until 7/14/2015, 34 eggs produced only 23 pups but still yielded 27 results. When an embryo died during development, if I noticed it soon enough through candling, the egg case was opened and the neonate sexed obtaining a result without an actual live pup. This method was used to sex embryos as young as 96 days. The results are shown in the table below.

	Male	Female	Mean incubation
Tank B (81°f)	7 (50%)	7 (50%)	118.9 days
Tank C (75°f)	6 (46.15%)	7 (53.85%)	176.3 days

Conclusion

While it would be beneficial to be able to decide the sex of unborn sharks ahead of time, it appears the temperature plays no part in determining the sex of the pups. This study is far from exhaustive, and perhaps with different individual adult sharks, or more dramatic temperatures one may obtain a different result. In our second group in particular, the temperature did not affect the sex of the sharks. The results of the warm and cool water group are almost identical, and evenly split between the genders. It was interesting to note how dramatically the temperature affected the incubation time, but it seems that was its only effect. I would have liked to have worked with a larger group of eggs for these trials, but by the time the second round was completed I had distributed 72 pups to facilities around the country. Being that our breeding pair was limited to 1.1, it seemed irresponsible to continue to produce genetically similar animals, and I was unwilling to euthanize shark pups in order to have a larger sample size.

Figure 1. one of the first pups hatched.

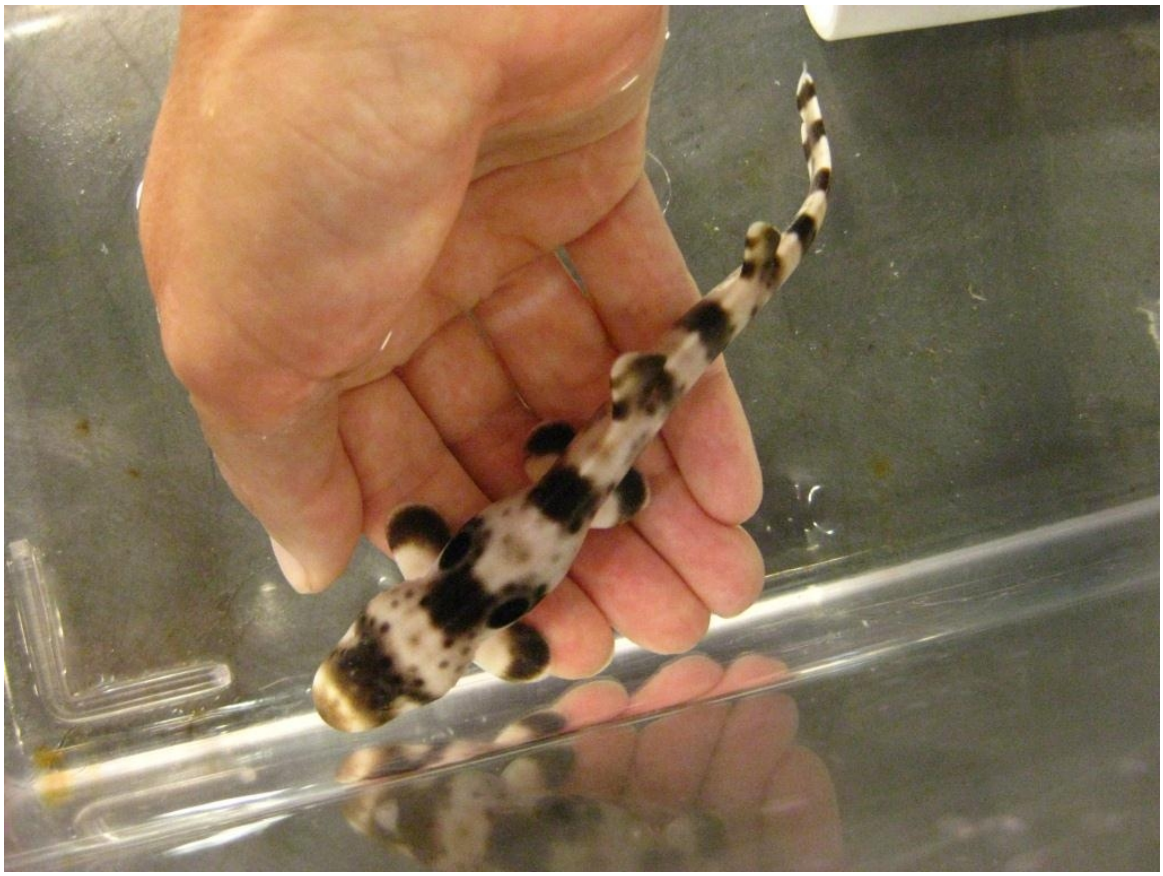

Figure 2.

Figure 3.

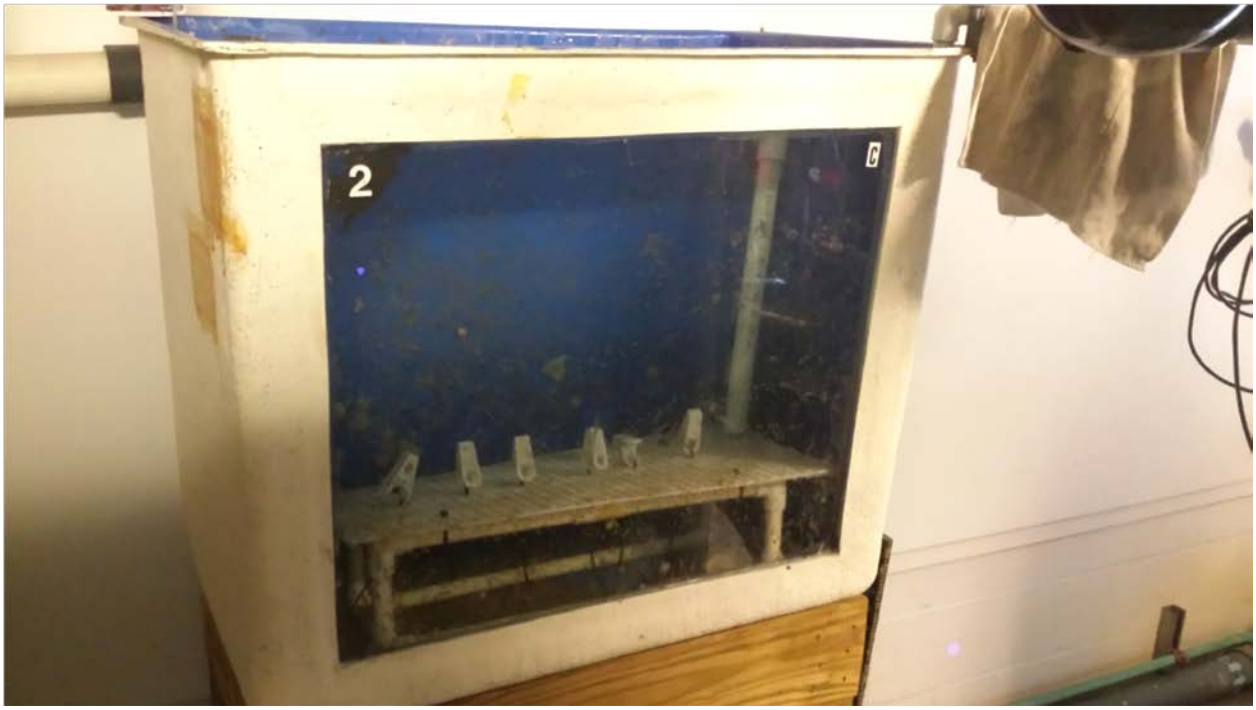

Figure 4. Initial egg holding tank plumbed into main exhibit (Tank A)

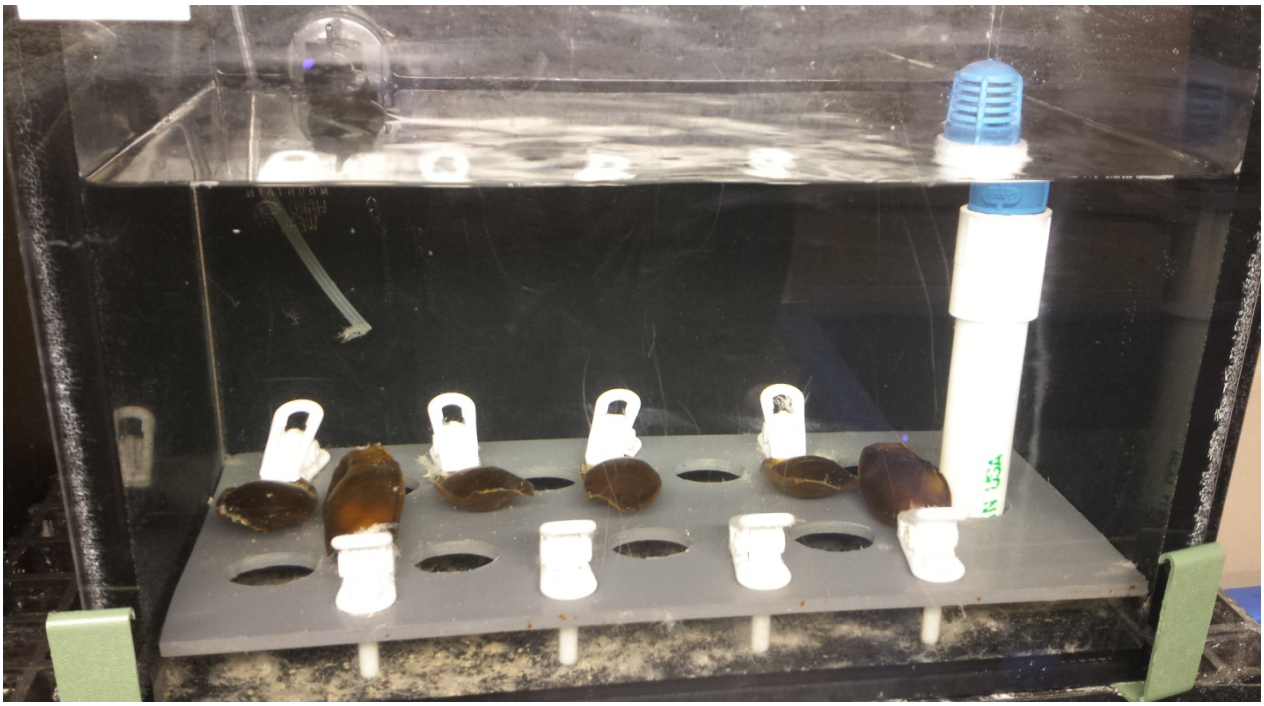

Figure 5. Warm water 10 gallon egg holding tank (Tank B)

A SIMPLER ELECTRIC EEL DETECTOR

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Children's Aquarium at Fair Park /Dallas Zoo Dallas, Texas

In the 2012 issue of Drum and Croaker, Markus Dernjatin wrote an article describing a simple electric eel detector. Here, I'd like to build on that article and suggest a few shortcuts.

Our electric eel detector system, while fashioned on Dernjatin's design, incorporates a couple items making the set up somewhat simpler. Two one meter stainless steel rod detectors are placed into the tank with PVC pipe sheaths to keep the eels from directly touching the detectors. We hooked these up along with a 0.01mF 200v capacitor in line to a ¼" microphone plug that is inserted into the microphone jack of a small amplifier [Pyle PTA4]. The sensitivity of the signal can be adjusted using the mic volume on the amplifier. A speaker can then be attached to the amplifier for sound, but the real innovation is that we also connect 12 volt LED lights directly to the speaker ports on the amplifier. Whenever the eels send out a pulse, the speaker pops and buzzes and the lights flash in response. With this design, a light control unit as described by Dernjatin is not needed. Then, we added a few refinements. While we have the lights flash with every pulse, we incorporated a button that the public pushes to hear the pops thus making the unit more interactive. In addition, we wired in a hidden switch that can be activated for continuous sound for use during education demonstrations, etc. Finally, a "pig tail" was added in order to attach a Christmas tree. Again you must use 12 volt LED Christmas lights.

Our system was designed and built entirely in house and has been working for a about a year. It is very popular with the public. The Christmas tree addition, while certainly not an original idea, is used during the holiday season and has been enjoyed by our visitors and has garnered local press attention. A view of our system in action including the Christmas tree can be found here.
https://youtu.be/KHW7KQRyABA

I wish to thank the folks at SEA LIFE Center in Helsinki, Finland for their generous assistance as I began setting up our detector.

CHLOROQUINE PHOSPHATE AND PRAZIQUANTAL AS A MEANS TO ERADICATE *Decacoytle floridana* IN SPOTTED EAGLE RAYS

Kelly Sowers; Ed. Latson DVM; Robert George DVM; Leah Neal

Ripley's Aquarium of Canada

Abstract

In an effort to maintain life expectancy and animal health in a managed care environment for spotted eagle rays (*Aetobatus narinari),* a new approach for treating a common monogenean, *Decacoytle floridana*, was implemented. Praziquantel and chloroquine phosphate have both been used in marine system for a variety of parasites. By utilizing these two chemical treatments in tandem, the eradication of the target monogeneans was apparently achieved. The positive results achieved with this method can lead to improved health conditions of managed spotted eagle ray collections in the future.

Introduction

Spotted eagle rays (*Aetobatus narinari*) are a dynamic addition to a tropical marine aquarium. While generally a docile animal that responds well to dive shows and hand feeding, issues with internal parasites as well as external monogenean infestations are fairly common and challenging. Eagle rays collected from the wild often arrive infested with a variety of monogeneans that can prove difficult to eradicate. *Decacoytle floridana* (Dfl) is one such parasite. It is a rapidly reproducing, species-specific monogenean that utilizes several hook-like anchors located on the opisthohaptor to pierce the gill tissue of the host (Noga, 2010). The parasite then feeds off of the host's gill tissue and blood. Although very small in size, the adult parasite can be very destructive. Large numbers of this monogenean can greatly affect the health of the host animal, causing substantial morbidity and, if untreated, mortalities. A spotted eagle ray known to be infested with Dfl was placed in a quarantine system with an existing population of eagle rays and treated with standard quarantine protocols of oral medications and praziquantel baths. The Dfl was reduced in number but not eliminated and the parasite continued to reproduce.

In an attempt to eradicate this parasite, a plan was created with the intent to interrupt the direct life-cycle of the monogenean by employing two medications; serial applications of praziquantel to remove adult worms and sustained immersion of chloroquine to inhibit oncomiracidium development. Dfl reproduces in an oviparous fashion in which numerous eggs are released into the water column. Like other monogeneans, Dfl ova may start to hatch after three weeks but may continue to hatch over prolonged periods of time. The hatchling is a ciliated, free-swimming oncomiracidium that attaches to a host on which it will stay for the rest of its life (Noga, 2010). Previous studies have shown that while the adult stage is affected by a common anthelmintic, praziquantel, treatment with praziquantel alone does not guarantee complete eradication. An additional drug was added to the treatment protocol which would focus on the ciliated stage of the monogenean. Chloroquine phosphate was selected because it has been used to successfully treat a variety of ciliated parasites (Hemdal, 2013). It has long been used as an antimalarial drug in humans and birds. The therapeutic index is narrow in humans and birds so monitoring for side effects was important to the safety of the animals.

While there is little or no information on how the spotted eagle rays would react to chloroquine alone or coupled with praziquantel, the following case study illustrates how eradication of *D. floridana* was apparently achieved.

Methods:

An extended treatment was designed using two different chemicals in tandem; praziquantel and chloroquine phosphate. The treatment was performed in a serious of steps:

The initial treatment was performed in a 3,500 gallon transport tank which was dosed with 5ppm praziquantel before introducing the rays. Three subadult eagle rays were immersed in this system for 30 hours. The animals were carefully monitored for any signs of stress. Water samples were taken from the transport tank after the bath was completed and the animals were moved out of system. Eggs and live adults in large numbers were found in samples obtained from the treatment tank water. A large percentage of the adult stages were found in a paralyzed state but showed signs of recovery as time went on. The eagle rays were not returned to their original tank, but instead placed into a tank that had not housed eagle rays or been exposed to this particular monogenean. All eagle rays successfully completed the bath with no apparent negative side effects. Behaviors and eating habits remained normal after removal from the treatment tank and being placed in the permanent 20 foot diameter tank. This tank also housed two juvenile cownose rays (*Rhinoptera bonasus*) and one adult atlantic ray (*Dasyatis sabina*).

Samples were taken of the untreated water to create a standard with which to monitor chloroquine levels during treatment. Using a HACH 5000 UV spectrophotometer, a standard curve of absorbency was created by passing a 329nm wavelength light through samples of known chloroquine concentrations (Hemdal, 2013). The concentration levels ranged from 0ppm, for untreated tank water, to 40ppm. The result was a set of relative values that allowed daily monitoring of chloroquine and the ability to dose and maintain the desired concentration in the treatment tank. Figure 1 illustrates the curve created.

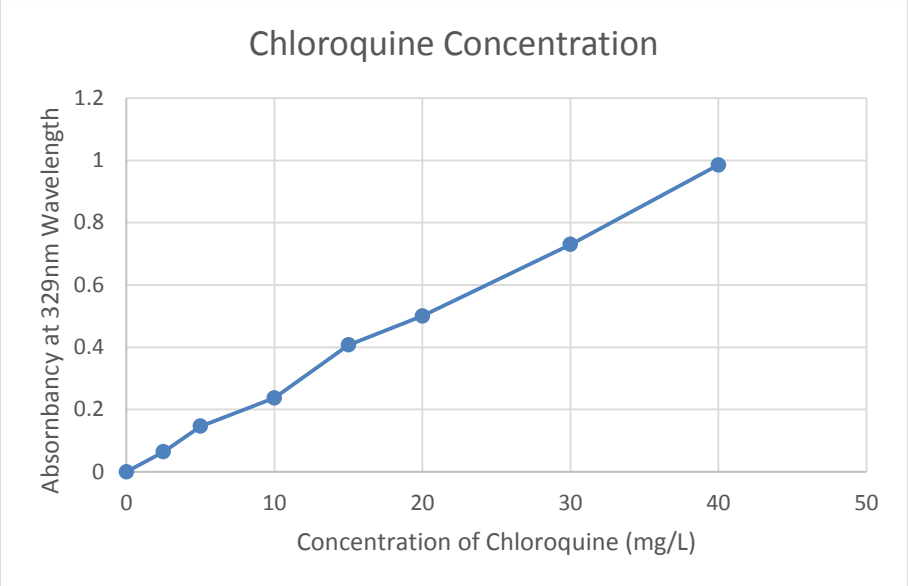

Figure 1. Chloroquine concentrations based on absorbency readouts utilizing a HACH 5000 spectrophotometer.

The 20’ diameter tank was dosed with 3ppm praziquantel and 3ppm chloroquine before animals were introduced to the system. Chloroquine levels were then increased slowly. Animals were observed closely for any signs of distress. By raising the concentration of chloroquine by 2ppm or 3ppm every other day, a final concentration of 10ppm was achieved in a week’s time. Employing the previously described standard curve, chloroquine additions were done whenever system concentrations dropped. The chloroquine was very stable and additions were only needed when replacing water lost in water changes.

Praziquantel was added into the system to a concentration of 3ppm. Praziquantel is metabolized by bacteria in the biofilter and is very short lived in systems that are treated frequently (Thomas, et.al. 2015). In order to maintain the therapeutic praziquantel levels in the short period between hatching and adulthood, the tank was treated every 5 days to prevent DFL from reaching the adult stage when they would begin to lay eggs. A total of twenty-six additions were added over the entire treatment period of 4 months.

In order to monitor the tank for the presence of the monogenean, water samples were taken multiple times a week. Additional samples were obtained by siphoning the bottom of the tank approximately an hour after each praziquantel addition. After trying a series of different sampling techniques, water samples taken from the fractionator and samples siphoned directly from the bottom of the tank proved to be the most effective at confirming the presence of monogeneans. Water samples were centrifuged to isolate debris and monogeneans. The centrifuged portions were then placed onto a microscope slide and examined. Both the eggs and the adult stages were easily identified at 10X magnification.

Monogeneans, in both egg and adult life stages, ceased to be detected after seven praziquantel additions and the chloroquine levels had been steady at 10ppm for twenty-eight days. Despite the apparent success, the treatments were continued for another 3 months. In total, 26 doses of praziquantel were added to the tank and the chloroquine was maintained at 10ppm for a total of 5 months.

After the treatment period, all praziquantel and chloroquine was removed via activated carbon, ozone injection, and water changes. All three eagle rays continued to behave and eat normally with no interruption in feeding. Water sampling frequency was reduced to once a week to monitor the tank post-treatment.

The final step was to attempt to verify that the monogeneans had been eradicated before moving the rays into the permanent exhibit space. To do this, the eagle rays received gill flushes utilizing a technique being used in other public aquarium settings. This procedure took place five months after the last praziquantel addition and 4 months after chloroquine levels were dropped below treatment levels. The rays were individually placed into a 225 gallon tank which had already been dosed with 50ppm MS-222 and equal parts sodium bicarbonate. The rays were then placed into tonic immobility to allow access to the gill slits. The set-up for the flushes included two five gallon buckets, one with system saltwater, the other with freshwater. Both buckets were dosed with 15ppm praziquantel. Using a small magnetic drive water pump attached to plastic airline tubing, praziquantel mixtures were flushed over the gills of the eagle rays. The airline tubing was inserted into each gill slit several times to flush the parasite into the

water column. After the flushing was complete, each animal was moved back into the main tank for recovery. Multiple samples were taken from the flushing water and examined using the same sampling techniques used to monitor the holding system.

Results:

During the initial 5ppm 30 hour praziquantel immersion, the monogeneans were affected but not completely removed. Sampling of the treatment tank water showed there was some success in getting the monogeneans to release their hold on the gills, but did not kill them. The majority of monogeneans had released their hold on the gills and were noted as being shriveled, but still were mobile, suggesting the praziquantel had only temporarily paralyzed them. Very soon after the animals went into the clean treatment system, eggs were being produced and found when water was sampled. Therefore, the initial 5ppm praziquantel was not successful in removing all of the adult worms. Some of the worms may have remained lodged in the gill crypts and revived when the rays were placed in the holding tank. Another possibility could be that unhatched eggs were been caught in gill filaments then later hatched, carrying on the life cycle.

The best source for water sampling was the fractionator. Foam was collected and liquefied by adding a small amount of tank water. These small liquefied samples were then centrifuged so the precipitate could be examined by microscope.

All of the eagle rays reacted well to the system treatments and the gill flushing. To monitor for diet, behavior, or energy level changes, these animals were observed daily. Throughout the entire trial, the eagle rays did not show any signs of stress or changes. Cownose rays (*Rhinoptera bonasus*) kept in the same system showed changes in swimming behavior and were moved out before the treatment was completed. Immediate recovery was noted and what aspect of the treatment they were not able to tolerate was not determined. The atlantic ray (*Dasyatis sabina*) remained in the treated system for the entire period and showed no negative effects from the treatment.

After performing the gill flush on the spotted eagle rays, all animals recovered immediately and resumed normal swimming and feeding behavior. Multiple samples were taken from the holding bins post-flush. No monogeneans of any life-stage were noted. Animals were then deemed as clean and fit to be moved onto exhibit.

Discussion:

Multiple praziquantal treatments administered over a prolonged period of time with short inter-treatment intervals along with prolonged chloroquine phosphate exposure appears to be an effective method for eliminating *Decacoytle floridana.* It is not clear which medication was the successful component of the treatment protocol or if it was the combination of drugs that eliminated the parasite.

While the eradication of the target monogenean *Decacoytle floridana* was apparently achieved, there are many aspects of the treatment that can still be explored. One such aspect is the effectiveness of each medication by itself.

Due to our small staff and time constraints, the presence of eggs was noted on a present or not-present basis. By utilizing more thorough counts of eggs, the gradual decline of the monogenean may be more accurately tracked.

Sampling of the permanent exhibit which the rays were transferred to has been done in an attempt to determine if the monogenean has been truly eradicated. No life stages of the monogenean have been found at the time of publishing. Continual sampling will be done over a long period to verify there is not a reinfestation.

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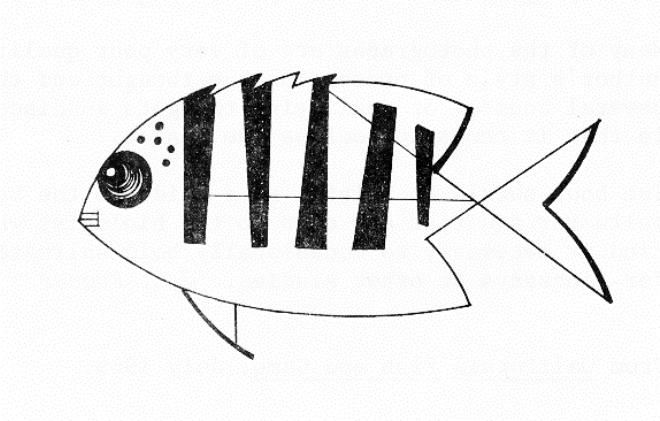
Personal Communications:

Much of the information and methods decided on were reached in verbal communications between the following:

- Dr. Robert George and Dennis Stoney. Discussion on utilizing the tandem approach in the chloroquine phosphate and praziquantel.
- Author and Jim Kinsler, Sea World. Sharing of Dr. DiRocco's (Sea World, Orlando) gill flushing method.



CULTURE METHODS OF THE GRASS SHRIMP, *Palaemonetes* sp.

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Abstract

Central Campus is a Regional Academy in Des Moines, Iowa that provides students unique and advanced opportunities to explore a career in Marine Science, among other fields. One such opportunity that the Marine Biology program recently took on was to culture the grass shrimp, *Palaemonetes* sp. The *Palaemonetes* culture system was initially started to provide live food items for specialized organisms in the lab that require live food, such as the leaf scorpion fish, *Taenionotus triacanthus,* and the painted frogfish, *Antennarius pictus*. After several 'trial-and-error' variations of culture systems, the final setup proved to be most effective at conserving space, providing good water quality, and maximizing hatchings. The current culture system has been up and running for six months and has produced approximately 400 additional grass shrimp. The success of this system has enabled the program to add additional organisms to its collection that require live food, such as the pyjama squid, *Sepioloidea lineolata*. This project, and similar projects like these in the program, highlight the hard work and innovation that students at Central Campus continue to show. To highlight the student experience at Central Campus, along with describing the culture system, first-hand accounts of the primary author (Walker) were added within the text.

Introduction

Central Campus is an advanced Regional Academy in Des Moines, Iowa. Students from surrounding high schools have an opportunity to attend the school, free of charge, and explore their interest in a number of career areas while also earning college credit. Central Campus offers a number of courses within the fields of Aviation, Construction, and Automotive to Fashion Design, Graphic Design, and Photography to Marine Science, Horticulture, and Nursing. The programs within Central Campus gives students from all backgrounds a strong foundation to begin their adult lives; it gives the students a nurturing environment that they are encouraged to explore their options and pursue them if they wish.

Within the Marine Science Department, students may take a class in either Aquarium Science or Marine Biology. These classes work closely together, by running different aspects of the 14,000-gallon saltwater lab that they share; although they do have separate classrooms where they learn the specifics of each trade. Generally, Aquarium Science students focus on aquaculture and the specific requirements for maintaining marine organisms in captivity. Marine Biology students focus on research projects and the diversity of marine life in the oceans. All of the students rotate through different tanks during the year, learning how to provide different husbandry techniques for a variety of marine organisms from jellyfish, sea horses, and sharks, to sting rays, nautiluses, and anglerfish. Overall the Marine Science classes are set up to thoroughly teach students how to excel in their choice of study. After taking one of the classes, students may apply to be a Summer Lab Manager or a Lab Assistant, both of which provide students

additional responsibilities and opportunities to pursue specific projects, such as setting up and maintaining a grass shrimp, *Palaemonetes* sp., culture system.

STUDENT POINT OF VIEW *– "I personally went through the year-long Marine Biology program, and at the end of the year I was given the opportunity to be one of three lab managers for the summer. I spent 5 days and an average of approximately 3.5 hours a week at the lab. While there, I shared the responsibility of maintaining water quality, feeding, and other jobs to keep the lab running over the break. I was solely in charge of the continuation of the grass shrimp breeding program that carried over into the beginning of the next school year, and is currently what I spend a majority of my time on at the lab, along with other small projects like testing water quality or feeding. When I first started working with the grass shrimp it was just another job, but it has turned into something that I am passionate about, and look forward to watching it succeed."*

Palaemonetes sp., grass shrimp, are found in shallow coastal waters, salt marshes, and seagrass beds, and live in both brackish and saltwater; they are most commonly found on the southern coast of the United States. These crustaceans are omnivorous, eating a range of plants, and small animals such as phytoplankton, detritus, and invertebrates. *Palaemonetes* play a large role in the ecology of the environment that they live in (Welsh 1975). Not only do they provide larger animals with a source of nutrition, but they also break down detritus and make it easier to be eaten by smaller organisms, thus starting the cycle over again. *Palaemonetes* have been used successfully as a prey item of post-hatchling cultures of cuttlefish (Richard 1971; Pascual 1978; Dominques et al. 2001a; Dominques et al. 2001b; Dominques et al. 2003). Prior to the use of *Palaemonetes*, live mysid shrimp, *Americamysis bahia*, are often used as the prey item for the first few weeks after hatching. However, this diet is not commercially viable as a food source due to the high costs (Sykes et al. 2006). Thus, the use of a single prey item throughout the culture of cuttlefish could reduce costs, labor, and increase culture rates (Sykes et al. 2006).

Having student partake in this project not only teaches them fundamentals of maintaining a proper habitat for these animals, but it also strengthens problem solving skills. During the process of creating a successful breeding program the students deal with challenges and problem that they must find a way to overcome to better the program. They learn how to work together and how to better communicate with others. One student is not always able to be at the lab caring for the animals, because of that it is important that he or she should be able to thoroughly explain how to care for the shrimp and to update on any changes that may have happened. Overall these shrimp play an engrossingly important role in the lab and how it changes.

Methodology

The current *Palaemonetes* culture system consists of eight 20 liter, white, plastic bins (35cm x 25cm x 25cm), Fig. 1. The white color allowed for relatively easy viewing of the hatchlings as well as detritus and waste. Each bin was filled with 18 l of artificial saltwater. These eight bins were used for hatching and grow-out. A bio-sponge filter system was used for each bin to maintain water quality. Eight smaller clear, plastic bins (15cm x 15cm x 15cm) with holes (~1cm in diameter) drilled into the sides and bottom.

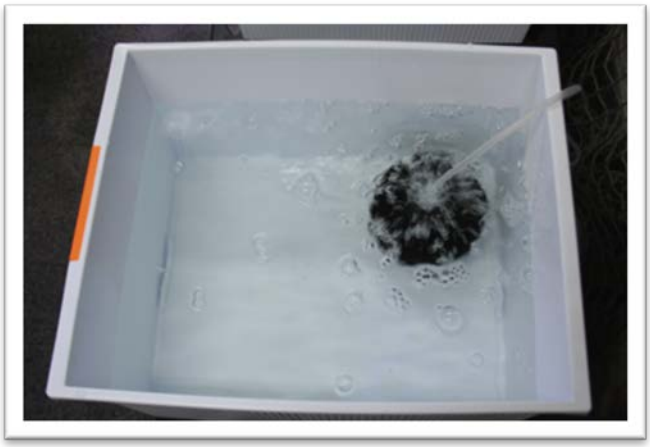

Figure 1. Large, white culture bin.

Figure 2. Gravid female *Palaemonetes*

A maximum of 10 gravid females (Fig. 2, 3) were removed from brood stock holding and placed in the smaller bin, which was then floated in the larger bin (Fig. 4). When the eggs hatched, the hatchlings could move through the drilled holes of the smaller bin and into the larger white bin – the holes were small enough that the females could not escape and potentially consume the hatchlings.

Figure 3. 10 gravid *Palaemonetes* in small bin.

Figure 4. Initial startup of culture system.

The gravid females were fed flake food daily and as the eggs hatched, *Artemia* nauplii was fed to the hatchlings. One table spoon of concentrated *Artemia* nauplii was diluted in 1 liter of artificial seawater. A total of 18ml of this mixture was then target fed to the areas of the bin that the hatchlings clustered in. A flashlight was also turned on prior to feedings to attract the hatchlings, as well as the *Artemia* nauplii, for more directed feedings and estimated counts. As the hatchlings grow, powdered flake food is added to their diet to transition off the *Artemia* nauplii and the diet will eventually consist of whole flakes when the juvenile grass shrimp attain a size of 1-2cm. The bins are labeled with different colors depending on what each bin should be fed and the age of the hatchlings within the bin (Fig. 5).

Figure 5. View of four culture bins setup with gravid females.

Water quality was maintained through the bio-sponge filter system and daily siphoning of waste with accompanying water changes. Salinity was checked daily and ranged from 31-38 ppt. The temperature range was 23-25 °C and was maintained by the labs ambient temperature control. Other parameters such as ammonia, nitrite, and nitrate, were checked as needed and were within acceptable standards.

Results

The program was started in the spring of 2016 and since then, approximately 200 grass shrimp have been added back into the brood stock after reaching adulthood. Generally, each group of 10 females has produced approximately 60-120 hatchlings. At the time of this publication, there are approximately 200 additional hatchlings in the culture bins.

Discussion

The initial *Palaemonetes* culture system started by utilizing recycled 2-liter plastic bottles. Each bottle housed a single egg-carrying female which was removed from the bottle after the eggs hatched. While the 2-liter bottles minimized space requirements and re-used bottles, the small water volume led to problems maintaining good water quality conditions. Soon after, the current plastic bins were used and proved to work much more effectively. The *Palaemonetes* culture system quickly evolved as different students participated in the project and new ideas were both unsuccessful and successful. The successful evolution of this culture system was really a prime example of the opportunities students have at Central Campus to troubleshoot problems on their own.

STUDENT POINT OF VIEW *– "The project started out very time consuming, but there is a large pay off, and makes it possible to explore new/different organisms. I would estimate that it takes at least an hour a day to maintain regularly and, if something special needs to happen it takes more time. Things that may take more time for instance would be: counting the shrimp to be returned back into brood stock, a large water change due to overfeeding, or even starting a*

new bin on top of the normal protocol can add a substantial amount of time. There is a learning curve to the project simply because there is a large number of things to learn, but after you are comfortable with the protocol the time moves quickly. Since this is an ongoing project it changes constantly, and we are always thinking of ways to work out the problems."

The amount of time required to maintain the culture system was magnified when training new students on the methods to maintain the system. Within a teaching environment, there will inevitably be a learning curve and alongside continuing to improve the output of the culture system, it was also necessary to continually improve the husbandry protocols to reduce the learning curve.

STUDENT POINT OF VIEW **– "***When I first started working with the grass shrimp I battled the way we had been feeding, it was inefficient, and in the end, it caused more work because the over feeding caused poor water quality. To maintain a decent water quality, we would have to do water changes quite often and, at that time the bins were bigger; this wasted a large amount of saltwater and food. To solve this problem, I pinpointed what each size of shrimp should be eating, and that cut down on wasted food. We also moved to using smaller bins; this does make it harder to control water quality but, it cuts down on wasted water because of the frequent water changes.*"

The initial diet of *Artemia* nauplii fed to the hatchlings not only ended up degrading water quality but also may have been the primary reason of several significant mortalities observed. On several occasions, 100-200 hatchlings were observed soon after the gravid female *Palaemonetes* were added to the culture bins but after 2-3 weeks, the numbers significantly declined – in some cases, all hatchlings were lost. Water quality parameters remained within acceptable parameters for most of the culture bins (the increased salinity in one bin may have resulted in mortality). Rather than water quality, poor diet was the next potential reason for the losses. After close observations, the bins still had large concentrations of the *Artemia* nauplii several days after their additions, at which point their nutritional value would be minimal. Thus, as new *Artemia* nauplii was added, the hatchling *Palaemonetes* consumed organisms closest to them, whether they be fresh or several days old. This may have resulted in problems progressing through the growth stages and eventual mortality. After these observations, the amount of *Artemia* nauplii fed was significantly reduced and the feedings were targeted to where the hatchlings grouped together. This change proved to be successful at raising hatchlings pass this stage.

STUDENT POINT OF VIEW *– "Overall my experience with breeding grass shrimp has been a very time consuming process that has taken a lot of trial and error to figure out, but by doing this I have found a process that works very well for me, and has cut the time I spend working on the shrimp in half, even with the expansion of the project. My experience with the grass shrimp has been more then worthwhile, and it has allowed the lab to move forward with obtaining new and exciting organisms."*

Conclusion

Without the ability to culture organisms like *Palaemonetes*, the program at Central Campus simply could not continue to grow and be able to bring in new organisms for the students to work with and learn about. The current culture system has provided the resources for the Marine Biology program to expand by providing a consistent source of live food items and reducing our reliance on continued life food shipments. In addition, we also believe this culture system can be repeated at other facilities to support their live food needs. Certainly, the challenges in any life food culture system include space requirements, maintenance times, and eventual output. When fully up and running, the 8-bin culture system should be able to produce an average of 800 *Palaemonetes* every two months. To support and continue to improve upon these results, smaller culture bins will be used to house single gravid females to improve our understanding of fecundity per female. During these trials, the diet provided to the hatchlings will also be varied to determine the most effective diet types and amount of food.

Sykes et al. (2006) raised *Sepia officinalis* through five consecutive generations on the sole diet of *Palaemonetes varians*. Although even the small grass shrimp were twice the size of hatchling *S. officinalis*, they were still consumed at this stage. The potential of using a different food source for hatchling cuttlefish will significantly increase future opportunities for additional facilities to exhibit cuttlefish, for both education and research. The reduced costs and labor associated with this prey item, as opposed to *Americamysis bahia*, also support large-scale aquaculture of cuttlefish for human consumption. Continuing this line of research, then, is critical to our understanding of the metabolic needs of cuttlefish which will help inform future conservation efforts.

STUDENT POINT OF VIEW *– "What I can say is that to this point we have found success in how we are breeding the grass shrimp. There are things about the project that still need to be changed in order for us to be satisfied with the yield we are getting, but we have a strong foothold to continue growing. While taking care of these shrimp I have discovered that there is really no way to perfect a changing process because the way you execute the process must change with it. Every time the process changes, problems occur that have to be fixed. This project has not only helped the lab grow, but it has taught me personally so many things; I have learned to identify what is causing a problem, and to come up with ideas on how to counteract the problems, it showed me how to manipulate water quality This project has showed me what being really dedicated to something means, it showed me that even when you work hard on something it doesn't mean that it will work out exactly how you wanted it to, and lastly it showed me what it feels like to struggle with something and overcome it."*

Acknowledgements

The authors would like to thank Des Moines Public Schools and Central Campus Regional Academy for providing the environment and opportunity to work on this project. Mr. Kirk Embree was essential at troubleshooting different ideas and solutions to problems along the way. The summer co-managers of the lab, Owen McGuffin and Katelynn Pierce, helped care for the culture system when H. Walker was away and also assisted with other lab duties. Noel, Mark and Gavin Walker helped take care of the shrimp when others were unavailable. Skylar Middle and Ross Thompson provided additional assistance during the summer. Josie Noland has started helping out during the fall semester and will be taking over the project after H. Walker graduates.

Last but not least all of the people who take care of the shrimp when I am not around, or the people who help with tasks that go unnoticed. Without these additional students the program would surely not be the success that it is today.

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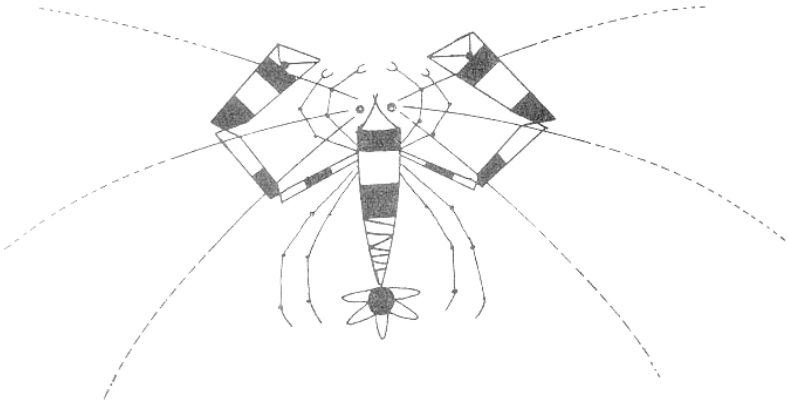

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Tuesday, March 22
Session 1: Sharks/Sand Tiger Shark Roundtable Discussion

The Legislative Conservation Status of the Sand Tiger Shark (*Carcharias taurus*)

Joe Choromanski
Ripley's Aquariums
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The current legislative conservation status of the sand tiger shark varies by population, ocean and country. The entire population is viewed as VUNERABLE under the IUCN Red List with certain distinct populations in the SW Atlantic and both coasts of Australia as CRITICALLY ENDANGERED. This brief presentation will provide additional details on their legislative protection status.

Development and Dissemination of Tools to Access Reproductive Status of Sand Tiger Sharks (*Carcharias taurus*) and Advance the Field of Assisted Reproduction in Sharks.

Jennifer Wyffels[1] Jennifer.Wyffels@sezarc.com,
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Nearly a quarter of all elasmobranchs, including the sand tiger shark, are listed in the IUCN Red List of Threatened Species. Despite a long history of husbandry for sand tiger sharks

dating back to the 1930's, reproduction under managed care has been mostly unsuccessful. In 2014, the Florida Aquarium and North Carolina Aquariums, joined the South-East Zoo Alliance for Reproduction & Conservation (SEZARC) to spearhead a growing consortium of AZA institutions to work on a holistic approach of ensuring propagation of sand tiger sharks in aquaria. Understanding the reproductive seasonality of sand tiger sharks under managed care is the foundation for reproductive success. Focusing on the male, enzyme-linked hormone assays (EIA) have been validated for testosterone and serial dilutions (range=neat to 1:128) of pooled plasma yielded displacement curves parallel to the standard curve and recovery of known amounts of testosterone (0.002 – 1.0 ng/ml) added to pools of diluted plasma (1:100,) was 107% (y=0.9691x-0.0003, r2=0.999). Sharks (n=7) were sampled quarterly (n=3) or once or twice a year (n=4) for semen collection. Spermic ejaculates (range ~7-105 ml) were obtained in all months sampled (Jan, March, April, May, June, July, October, and December) but motile sperm were obtained only in March (n=1), April (n=2), May (n=2) and June (n=1) supporting seasonality for semen production. Additionally, semen from males under managed care that were collected in April and May were not able to be collected within 30 days suggesting a possible finite annual semen production period. Aquarium and husbandry professionals have committed their institutional knowledge, experience, and resources to enable this highly collaborative multi-institutional reproductive research with the goal of rapidly generating and disseminating information to advance the field of assisted reproductive technology for these and other shark species under managed care.

Botox as a Possible Treatment for Spinal Problems in Sand Tiger Sharks (*Carcharias taurus*)

Dr. Robert Jones and Dr. Sam Gilchrist
The Aquarium Vet
Rob@theaquariumvet.com.au

Spinal problems (including scoliosis and kyphosis) are relatively common problems in Sand Tiger (*Carcharias taurus*) sharks maintained in captivity. A novel treatment using Botox has been trialed in one shark at the Manly SeaLife Sanctuary, Australia, with promising results over the past three years. The rationale for using Botox and a discussion of the procedure and outcome will be discussed.

Best of RAW Winner

Update on the Development of Assisted Reproduction Technologies in Elasmobranchs and the Wild Shark Rescues

Dr. Robert Jones and Dr. Jon Daly
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Assisted reproductive technologies (ARTs) include sperm collection and quality assessment, sperm cryopreservation, artificial insemination, and monitoring female reproductive condition and gestation via ultrasound. As a greater emphasis is placed on self-sustaining aquarium populations, ARTs will become an increasingly important component of breeding programs for elasmobranchs in aquaria. Ongoing research at Sea Life Melbourne Aquarium,

Australia, aims to create a basis for future use of ARTs in elasmobranchs in aquaria worldwide. This is to ensure sustainable captive populations of elasmobranchs, as well as having the potential for conservation of species in the future. An update on the current status of ARTs and future areas of research will be discussed. I will also update on the wild Sand Tiger (*Carcharias taurus*) shark rescues that are occurring in Australia.

New York Aquarium's New York Seascape Program and Sand Tiger Shark (*Carcharias taurus*) Field Research in the Great South Bay

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The 16,000 square miles of ocean from Montauk, New York, to Cape May, New Jersey, are an ecological treasure trove, providing critical migration routes for globally threatened animals, including sea turtles, whales and sharks, as well as nursery grounds and critical habitat for hundreds of species. These are also among the busiest waters in the world, with over 22 million people along the coastline. The main goal of the Wildlife Conservation Societies New York Seascape Program is to ensure a safe place for marine wildlife among the many competing human uses including shipping, energy development, and commercial and recreational fishing. One of the specific strategies to obtain this goal is to conduct field research; this research is conducted in order to understand the migratory and habitat use of key species. The main research project being carried out in the Great South Bay of Long Island is the equipping sand tiger sharks with acoustic tags. Between 2012 and 2015 27 sand tiger sharks have been tagged and through this effort an important nursery ground has been identified. The outcome of this project and the many others being conducted by the New York Seascape Program will be highlighted in the interruptive graphics in the New York Aquarium's new Ocean Wonders: Shark! exhibit.

Sand Tiger Sharks (*Carcharias taurus*) and The Graveyard of the Atlantic: Understanding the Role of this Critical Habitat.

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Internal Acoustic Transceivers Reveal the Annual Social Network Patterns in a Coastal Predator

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Sand Tigers (*Carcharias taurus*) are large apex predators resident in the coastal ocean along the Eastern US Coast. Although Delaware Bay and surrounding coastal waters are known

summer “hot spots” for Sand Tigers, our understanding of their seasonal movements is less well known. Since 2007, we have implanted more than 300 VEMCO acoustic transmitters in Sand Tigers, which have been detected from Cape Canaveral, Florida to Long Island, New York by collaborators in the Atlantic Cooperative Telemetry (ACT) Network. During the summer of 2012, 20 Sand Tigers were implanted with VEMCO Mobile Transceivers (VMTs), which are capable of both transmitting and receiving coded acoustic pings. To date, two of the 20 sharks have been recaptured, and their VMTs recovered. VMTs recorded detections of 350 individuals, from 8 different species. We analyzed their intra- and interspecific social network, which allowed us to reconstruct the approximate locations of Sand Tigers throughout the year. Changes in the interspecific population dynamics throughout the year revealed evidence of fission-fusion social behavior, which is common in mammals, but rarely documented in non-mammalian species. This project is a unique look at the social network of an apex predator and is a useful model for studies quantifying the social structures of marine animals. In addition, understanding how the aggregations of this species changes (in terms of sex and size class segregation) on spatiotemporal scales is critical for effective protection of the species and will be useful as managers develop conservation plans along the East Coast.

Best of RAW Winner

Call for Sand Tiger Shark (*Carcharias taurus*) Data to Analyze Captive Health and Potential Reproductive Cues

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In December 2015, the first sand tiger shark reproduction workshop was held at the White Oak Conservation Center. The workshop brought together aquarium personnel and researchers to create a vision and strategy for sustainable sand tiger shark populations under managed care and in the wild. During the meeting the need for increased information sharing about captive care, reproductive techniques, and wild populations was recognized as a vital conservation tool.

With numerous US facilities keeping sand tiger sharks for public display, the aquarium industry is poised to contribute on a large scale to sand tiger shark conservation in the face of declining wild populations, as well as sustainability and optimal husbandry for the display population. Many institutions are joining with the South-East Zoo Alliance for Reproduction and Conservation (SEZARC) in the process of developing reproductive techniques on adult sand tigers in captivity, including semen collection, preservation, and artificial insemination.

One vital component of this project is to identify parameters that may support successful captive reproduction and rearing of this species. To this end, the participants of the working groups would like to call on facilities caring for sand tiger sharks to contribute information on husbandry practices, behavior, nutrition, veterinary care, and outcomes. In order to create a comprehensive set of data that will allow researchers to compare variables on a large scale, we hope to collect holistic information from the time of animal collection to postmortem examination. We will present the framework for the data that is requested as well as a discussion of how the data will be collected, stored, and utilized for scientific gain.

Tuesday, March 22
Session 2: Husbandry

Globs in Globes: Thinking Outside the Kriesel

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Pioneering new jellyfish husbandry methods and liberal knowledge exchange have led the way toward jellies becoming a staple in many of the world's zoos and aquariums. Kreisels are often the go-to tanks for displaying jellies, but kreisels aren't the only tool in the shed. As it turns out some jellyfish do really well in spherical tanks resembling traditional fish bowls. Even more imaginatively, spheres can be plumbed such that the water leaves the tank through the top, cascading in gentle waves down the outsides of the tank, while the jellies remain splendidly inside. In this presentation I will take you on a transparent journey from seeing an idea for the first time in a faraway land, to bringing the concept to my new home aquarium, and refining the idea to suit. We will discuss the nuts and bolts of what has worked, what hasn't, and where we are now in the development of the jelly-sphere.

Best of RAW Winner

Care of the Endangered Copper Redhorse (*Moxostoma hubbsi*) at the Aquarium du Quebec

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The range of the copper redhorse (*Moxostoma hubbsi*), a critically endangered species of Catostomidae, is limited within a few rivers of the province of Quebec, Canada. Only a few hundred individuals are believed to be left of this species. A restoration program, involving re-stocking of hatchery produced fingerlings, was initiated by the provincial wildlife agency in collaboration with different partners including the Aquarium du Quebec. In 2009 a group of fingerlings were moved to the Aquarium for growth studies and future display as part of an educational exhibit on this conservation program. Very high mortalities, vertebral deformities and myopathy lead to the death or euthanasia of all of the fish within a year. In 2012 another group of fingerlings was obtained and a new diet and protocol was implemented. Following the detection of vertebral deformities in a few of the larger individuals the vitamin supplementation was increased (total of ~1000 IU of vitamin E and 2000mg of vitamin C per kg of food.) A decrease in the growth rate of the fish was also achieved by mimicking natural seasonal fluctuations in water temperatures. Following these changes, no new cases of scoliosis were observed and three years later a group of fingerlings are still alive and doing well. We are currently testing a pelleted diet (Vitalis, World Feeds) fed at 1% of the biomass daily as an alternative to our test diet.

Humphead Napoleon Wrasse (*Cheilinus undulatus*): Natural History and Husbandry of the Iconic Reef Giant

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"A rose by any other name…" They've been called many different names; Maori wrasse, humphead wrasse, giant wrasse, Napoleon wrasse, Wally, but whatever the name, *Cheilinus undulatus* is undeniably one of the most spectacular and beloved fishes in the ocean. In aquaria, they are equally admired and are quite possibly the perfect fish; a beautiful, intelligent, showy and impressive creature that can inspire personal connections with any guest that encounters them. This bond forms a lasting platform to engender a conservation ethos that few other species can achieve.

Is there potential to breed this species in captivity in public aquaria? In order to better understand these amazing fish, it's important to consider and know more about their day to day lives on the reef.

This presentation will reveal some of the unique and little known habits, hang-outs and habitats of this species, from what they eat and where they sleep to how they socialize and reproduce. There will be exclusive video footage of wild spawning aggregations as well as sneak peaks into the homes and holes of humphead Napoleon wrasse amongst the coral habitats of the Great Barrier Reef and Coral Sea. In addition to their natural history, the presentation will also share how these large wrasses make their way to public aquaria; including how they are collected, handled and treated before being transported to various locations around the world. This presentation will advance the knowledge about the wild lifestyles of these incredible animals. It will educate aquarists about their best post collection care in hopes that husbandry and display of these iconic reef giants in public aquaria continues to improve.

Spinal Injuries in Green Moray Eels (*Gymnothorax funebris*)

Dr. Emily F. Christiansen DVM, Dr. Shane Boylan DVM, and Dr. Craig A. Harms DVM
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Green moray eels are common display animals in public aquaria, especially in large marine exhibits. Due to long spinal columns and a tendency to inhabit caves and tunnels where movements are restricted, they may be particularly prone to spinal injuries. We report 3 cases of spinal injuries in captive eels with very different presentations and outcomes.

Case 1 showed acute onset of discoloration and necrosis of the distal tail that progressed over several days, eventually resulting in amputation. Initially, the cause of the necrosis was unclear, but radiographs several years later showed a proliferative bony lesion of the spine consistent with a prior fracture just cranial to the amputation site. The eel recovered well, and is thriving 5 years later.

Case 2 exhibited a slower development of an abnormal curve to the cranial body and limited swimming behavior. Radiographs and CT scan indicated compression of several cervical vertebrae cranial to the swim bladder. The eel was treated with Adequan® and cold laser therapy with substantial improvement. Over approximately 2 years, it developed recurrent swim bladder over inflation and positive buoyancy and was eventually euthanized.

Case 3 presented with respiratory distress and evidence of acute blood loss into the coelomic cavity, with internal trauma initially suspected. Radiographs showed a compressive spinal lesion affecting several vertebrae adjacent to the swim bladder, and neurologic function in the caudal body was abnormal. This eel partially responded to supportive care and steroid therapy, but did not fully recover and died following approximately 6 months of treatment.

Individualized Symbol Based Target Training
of Five Yellow Stingrays (*Urobatis jamaicensis*)

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A long-term training regimen for the five yellow stingrays (*Urobatis jamaicensis*) in the Aquarium Science Program facility's 2,500-gallon tropical elasmobranch holding system was initiated in October 2015. Five unique shapes on PVC poles are being utilized to target train the stingrays, with the end-goal of target recognition independent of the animal's location in the tank. In the project's early stages, the stingrays associate the presence of targets with the occurrence of a feeding session, but not food distribution. This long-term training regimen will help alleviate aggressive behaviors that the stingrays display during feeding sessions, and assist in the record keeping of each individual animal. Individualized target training is discussed and compared to generalized target training, which is being conducted on four catsharks and long tail carpet sharks in a tank also a part of the system.

Wednesday, March 23

Session 1: Conservation and Propagation

Practical Techniques for Free-range *Mobula hypostoma* Health Assessments

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In recent years' new species, such as *Mobula hypostoma*, have been appearing in aquaria worldwide. In order to become more knowledgeable on their general health and nutritional needs, we need to understand the health/condition of their free-ranging counterparts. We can apply the information gathered from ultrasound, blood collection, gastric lavage, and body measurements to help aquariums maintain healthier specimens.

Shark Ray (*Rhina ancylostoma*) Breeding – What a Difference 711 Days Makes!

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In late 2015, Newport Aquarium confirmed that its two female shark rays (*Rhina ancylostoma*) were pregnant. On 5 January 2016, the older female "Sweet Pea" gave birth 711 days after her first litter in January 2014. This long period between the 2014 and 2016 births provided Newport Aquarium biologists ample time to review and analyze the first unsuccessful experience with neonate shark rays. The knowledge gained from the first group of pups was the foundation for a new approach in caring for neonate shark rays, one that has proven to be successful to date. The presentation will detail the husbandry changes made by the biologists for the second group of neonates, as well as discuss the second female's pregnancy that did not reach full term.

Best of RAW Winner

Bonnethead Shark (*Sphyrna tiburo)* Husbandry: Challenges and Successes from Parturition to Quarantine

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On August 7, 2014, with the collaborative efforts for wild capture with South Carolina Aquarium (SCAq), the New England Aquarium (NEAq) became home to two gravid adult female bonnethead sharks (*Sphyrna tiburo*). The females were housed in a 30' L x 15' W x 4' D oval tank. Sedated exams were performed shortly after acquisition to remove hooks and confirm pregnancy for both individuals via ultrasound. Parturition occurred on November 2, 2014 and November 15, 2014 respectively. The first bonnethead birth resulted in 7 healthy pups (6 females, 1 male), whereas the second pupping event two weeks later resulted in 8 pups, 5 of which were stillborn and 3 that died shortly after birth. Sixteen months of husbandry and veterinary care for the adult females, pre and post parturition, and 13 months of care for the bonnethead pups are outlined, including the transition of the two adult females to exhibit in the NEAq's Caribbean Reef Exhibit (Giant Ocean Tank).

Almost Three Decades of Husbandry of Bonnethead Shark (*Sphyrna tiburo*) Pups

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Dynasty Marine Associates, Inc. has been rearing captive born bonnethead shark (*Sphyrna tiburo*) pups from wild caught female bonnethead sharks since 1989. The Dynasty Marine Transport Team made the very first successful air shipment of any hammerhead shark species by shipping 10 captive born bonnethead pups to the Tokyo Sea Life Park for their

opening in 1989. This achievement was followed by the very first air transport of any large hammerhead species, in this case *Sphyrna lewini*, to Beijing in 1998. To acquire broodstock, we access wild caught pregnant females because the Dynasty Marine Husbandry Facility has no room nor budget to maintain a reproductively active colony of bonnethead sharks. During this time, we have enjoyed many successes and encountered some highly vexing problems. This presentation will discuss the techniques used in the husbandry of these animals including feeding, prophylactic treatments (using Trichlorfon, (Dylox), Praziquantel, Dimilin and several antibiotics). Most of the diseases and problems that have been found whilst rearing more than 1000 pups during almost 3 decades will be discussed. We will present brief video clips of bonnethead pups exhibiting the most frequent abnormal behavioral swim patterns associated with the hypothetical neurological diseases affecting these captive born sharks. Further we will give related basic statistical information for some of the most recent year classes, indicating the incidence and evolution of these diseases during the first 6 months of holding after birth.

Tropical East Atlantic Biodiversity

João Correia
Flying Sharks

Commensal Relationship of the Banggai Cardinalfish (*Pterapogon kauderni*) with Host Sea Anemones

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Endemic to the Banggai Archipeligo, Eastern Indonesia, the Banggai Cardinalfish (*Pterapogon kauderni*) is a heavily exploited species popular within the aquarium industry. As of 2007, this species is listed as Endangered on the IUCN Red List and is currently under review for its inclusion in the Endangered Species Act, due to its extreme restrictive geographic distribution, low productivity, high genetic structure, overexploitation, and threats to habitats and microhabitats. For its protection, *P. kauderni* relies on its commenstalistic association with living benthic invertebrates, mainly sea urchins, corals, and anemones; these commensal relationships are essential for its survival at all life stages. In the wild, *P. kauderni* has been observed in close proximity with tentacles of anemones, seemingly without triggering discharge of nematocysts or a feeding reaction from the hosts, including newly released juveniles. A laboratory experiment was carried out to investigate if *P. kauderni* possess an 'innate' protection against nematocyst discharge and/or toxins of its various host anemones, in particular newly released juveniles and brooding males. A group of fifteen captive-bred *P. kauderni* were purchased to ensure no prior sensory or chemical exposure to anemones, and reproduction occurred in isolation in a fish-only breeding system. Brooding males and newly released juveniles were then introduced to two known host anemones species in an anemone holding system using a forced contact method to observe behaviors of subject fish and anemones upon initial contact. Results showed minimal stinging response and 100% survivability of fish tested. It is concluded that the protection exhibited by *P. kauderni* against nematocyst discharge is likely innate. Further studies will aim to determine the physiology of *P. kauderni's* protection against the defensive and predatory

responses of sea anemones, and experimentation including sea urchins (*Diadema sp.)* and host anemones is being designed to give insight on microhabitat selection by *P. kauderni* juveniles.

‘Cowbird’ of the Sea, Lazy Parents, or Just Coincidence?
Rearing the Planehead Filefish (*Stephanolepis hispidus*)

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On June 7, 2015, a substrate nest dusted with demersal eggs was carefully extracted from the New England Aquarium’s (NEAq) largest exhibit, the Caribbean Reef Exhibit, or ‘Giant Ocean Tank.’ Since divers observed the blue chromis (*Chromis cyanea*) defending the collection site, the eggs were then promptly transported to the NEAq’s Animal Care Center in Quincy, Mass, in hopes of rearing them as a focus species under the institutional larval fishes sustainability project. However, about a week later, the story began to take shape: at 7 dph, a prominent dorsal spine became oddly apparent. With the beginnings of ventral spine by 12 dph, it was undeniably evident that these were not blue chromis larvae, but filefish! Five species of Monacanthidae were exhibited in the ‘Giant Ocean Tank’ at this time, but it wasn’t until 65 dph when it was feasible to count fin rays and positively identify them as *Stephanolepis hispidus*.

Eat this, Not that! Raising Aquarium Animals to be Responsible Seafood Consumers

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The New England Aquarium’s Conservation Department works with the Aquarium’s Animal Care Team to regularly assess the source fisheries of the fishes and invertebrates they feed to the collection animals. Most of these fishes and invertebrates are ‘forage’ species, which have unique life-history characteristics and play a vital role in the food web, feeding the populations many marine animals in the wild. The internal assessments focus on the impacts these fisheries have on the surrounding environment by evaluating the following: the health of the target species population; the effectiveness of the way the fisheries are managed; the amount and composition of bycatch in the fisheries, whether or not they interact with protected, threatened, or endangered species; and the extent of fishing gears’ contact with the bottom habitat. The results of the assessments are used in conjunction with animal nutritional and health needs, taste, size, and texture preferences, and other important factors to distinguish between different species choices and formulate recommendations for alternative species (including aquacultured species), where applicable. To date, the Animal Care Team has tried new products, including farmed trout and invasive green crabs, switched some sources to less impactful fisheries, including squid and smelt, and committed to diversifying food items for the animals.

Small Scale Aquaculture, What Does it Take?

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There are many reasons for breeding animals at institutions rather than sourcing through wild collection. Some of those justifications include sourcing cost, limited availability of endangered species, and poor shipping survival. Many facilities have a large exhibit in which their inhabitants naturally reproduce but do not attempt to rear because of concerns of space and time commitment. I will give an overview of how we, at the Columbus Zoo and Aquarium, have utilized opportunistic spawning events to rear animals in house how much time it really takes. I will breakdown the daily tasks it takes to rear fish and invertebrates starting with live food cultures and maintenance of larval rearing systems without overloading one's time commitments.

A Pilot Study of the Effect of Gut Loading live *Artemia sp.* With Probiotics on Survival During the Pelagic Phase of Juvenile *Hippocampus erectus.*

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Seahorse aquaculture is continuing to grow due to high global demand for live and dried specimens. However, several bottlenecks in culture exist with catastrophic losses often occurring during the first few weeks of rearing (CITE SOURCE). Several studies have indicated higher survival rates and increased growth in certain fish species when adding probiotics to the larval diet (Kozasa 1986). The purpose of this pilot study was to examine what if any effects would be observed when feeding Lined Seahorses (*Hippocampus erectus*) enriched A*rtemia nauplii* gut loaded with probiotics from 0 to 8-11 Days Post Release (DPR). Specifically, I examined the INVE product Sanolife MIC-F containing the bacteria *Bacillus subtilis, Bacillus licheniformis* and *Bacillus pumilu* (Blundell 2008). Two broods of *H. erectus* juveniles (n= 206, n= 223) born over a three-day period were distributed evenly into two 38L tanks. Both cohorts received enriched 36HR post hatch *Artemia metanauplii*, however only one tank received probiotic addition to the feed. Preliminary results indicate that probiotic gut loading of *Artemia nauplii* may result in deleterious positive buoyancy, possibly from increased bacterial activity in the gut. Survival rates between the two tanks varied significantly with the probiotic treatment exhibiting 62% survival and the non-probiotic treatment exhibiting a 77% survival to 8-11 DPR. The data from this preliminary trial indicate that the use of probiotics in Day 0 post release *Hippocampus erectus* juveniles may contribute to positive buoyancy leading the fry to become trapped at the water surface meniscus, resulting in increased juvenile mortality. I suggest further studies on the use of probiotics in seahorse culture to examine this problem. This study contributes to the potential use of probiotics in seahorse culture. To the best of my knowledge this is the first quantitative trial on the use of probiotic enriched feed in Day 0 post release juvenile *H. erectus.*

Best of RAW Winner

Smells like Ctene Spirit: Methods for Culturing the Comb Jellies *(Ctenophora) Mnemiopsis spp., Pleurobracia bachei,* and *Bolinopsis infundibulum*.

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Comb jellies (Ctenophora) are popular aquarium exhibit animals due to their unique body plans, translucence, and the prismatic effect of their cilia on exhibit lighting. Acquiring them for exhibits can be challenging, however, as they are not always available from wild sources, and historically they've been impossible to culture. Only *Mnemiopsis spp*. has been cultured by a small number of scientific institutions. While literature regarding spawing and developing embryos is abundant, information outlining post-hatch development and successful culturing is currently lacking. Therefore, there is a great need within the aquarium industry for developing ctenophore husbandry protocols.

We will present culturing techniques for three types of comb jelly; *Mnemiopsis spp.*, which is the most common ctenophore genus exhibited in aquariums, and two species local to Monterey Bay: *Pleurobrachia bachei* and *Bolinopsis infundibulum*. We will discuss tank set-ups, feeding strategies, and the water parameters needed to grow and spawn successive generations of comb jellies.

Best of RAW Winner

Mariculture of *Diadema antillarum* for Reef Restoration

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Populations of *Diadema antillarum*, the long spined sea urchin, were decimated throughout the Florida Keys, the Bahamas, and Caribbean waters in 1983 after an unknown pathogen killed up to 98 percent of this species from the coral reefs. It has been shown that the die-off of this keystone herbivore corresponded with the loss of coral, including *Acropora cervicornis*, due to algal overgrowth.

Between 2008-2012 Mr. Martin Moe and Dr. David Vaughn made great advancements in deciphering the requirements for successfully culturing Diadema for reef restoration. These achievements included keeping fecund adults in breeding condition, spawning, maintaining larvae from spawn to settling, including settling larvae under the appropriate cues and metamorphosis into juveniles. Despite protocol rigor, they have not been able to repeat this process since 2012.

The objective of this research was to work with Moe to see if we can replicate the early success and in turn, determine the reasons that could have caused the 2012 and 2015 failures.

In three independent larval rearing trials since August 2015, we have been running trials at our Apollo Beach facility while Moe continued his work in the Florida Keys. Using identical

protocols and local water and algae, we were able to successfully spawn, develop and settle Diadema from the second trial at Apollo Beach.

We will discuss the techniques and methods used to perform these trials, as well as compare and contrast the differences between Florida Keys and Apollo Beach to identify potential confounding factors.

Identification and correction of the factors that are preventing consistent rearing consistent rearing success will allow development of a Diadema mariculture project for restoration to reefs.

Aussie Masters: Sustainable Coral Collections from Pristine Reefs

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Some corals are so extraordinary in their color, quality and uniqueness they have earned a special name; they're known as "master corals". Where they come from in Australia is special too; the pristine waters of the world heritage listed Great Barrier Reef. The Great Barrier Reef and adjoining Coral Sea have long been known as the source of some of the best marine fishes and invertebrates in the world, and with the growing popularity of living coral reef aquaria, the reputation has grown. This presentation will provide some insight into how and why the specimens coming from this region are so incredible and different.

Comprehensive video and graphics of the areas in which some of the most desired iconic Aussie corals, like *Acropora anthocercis* ("Tiera del Fuego"), *Acropora microlados* ("strawberry shortcake") and *Acropora echinata* ("blue bottlebrush"), live and thrive will be shown. Also included will be rare footage of the inter-reef zone depicting the preferred habitat of enigmatic species like Acanthophyllia, Trachyphyllia and Catalaphyllia. From the incredible habitats of these corals, the ocean environment will also be considered. Comprehensive analysis of the water found in the immediate proximity where these corals are collected provides an exciting and logical step in the development of coral husbandry into the future. The results of preliminary tests will be shared and provide surprising results that may revolutionize the art of coral keeping.

In addition to information about the wild environment of these specimens, the sustainable collection methodology will be revealed to further the understanding of fishery management, husbandry and infrastructure required to facilitate the shortest of live coral supply chains. All of these components in combination are a must in order for master corals to be displayed in aquaria.

Fuzzy Wuzzy was a…. Nautilus! Rediscovering a Lost Species (*Allonautilus scrobiculatus*)

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Nautiluses are disappearing. Nautiluses are disappearing because humans are mining them to extinction. Humans are mining nautiluses to extinction so they can have their shell, or a necklace, or an earring. Humans are loving nautiluses to death. Nautiluses are dying because of this love. It's time for humans to love the right way. *Allonautilus scrobiculatus* is the new nautilus on the scene. Also called the fuzzy nautilus, *Allonautilus* represents a new genera of nautiloid evolution in a couple million years. The whole idea of nautiluses being 'living fossils' is being challenged. Nautiluses are highly evolved cephalopods adapting to their environment just like anything other species. Papua New Guinea represents the only confirmed place where *Allonautilus scrobiculatus* exists. To top it off, *A. scrobiculatus* exists sympatrically with *Nautilus pompilius*. Papua New Guinea also represents an area of the world ripe for expansion and industrialization, evidenced by recent mining operations in the deep sea already being approved. What could this mean for the fuzzy, wuzzy nautilus? What could this mean for the not so fuzzy, wuzzy nautilus? What could this mean for the deep sea ecosystem as a whole? What could this mean for Papua New Guinea? The World? In August 2015, our team traveled to Papua New Guinea to address these questions. Working with the local Mbunai tribe, we used a combination of methods, including underwater video, ultrasonic transmitters, and genetic analyses, to survey *Allonautilus*, *Nautilus*, and the deep sea ecosystem they share. What we were left with was hope, trepidation, thankfulness, and fear. The history of nautiluses shows us that they have survived for hundreds of millions of years, through all five mass extinctions. The future is less clear. How will humans continue to love nautiluses? How will you love nautiluses in the future?

Double Your Goodeids, Double Your Fun: Collaborating with Students to Increase Breeding Efforts and Promote Goodeid Conservation

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Nearly all goodeid species are in trouble in the wild, and according to the IUCN, roughly half are considered threatened or extinct. In addition to AZA FFTAG participants, there are a number of organizations like the American Livebearer Association (ALA) and the Goodeid Working Group (GWG) that are leading the charge in goodeid conservation; despite these efforts, these species are still underrepresented within the industry. Past RAW presentations and FFTAG meeting discussions have stressed that goodeids are excellent service aisle species, but many institutions simply lack the time and space required to maintain a population. I contend that institutions can still get involved in these efforts, even if they lack space for a dedicated goodeid holding system.

Local schools with "service aisle space" and an interest in aquatic conservation can be an excellent resource to assist public aquariums in their goodeid breeding efforts. Giving students

real-world, hands-on conservation experience can boost support for aquariums while encouraging students to pursue the hobby and potentially a career in the aquarium industry. Through this collaboration, aquariums can both promote goodeid conservation and spark the interest of the next generation of aquarists.

In this presentation, we will examine how students benefit from these hands-on programs and some examples of student work in goodeid conservation. Additionally, we will look at Audubon's work with the White Trim Goodeid (*Zoogoneticus tequila*), a species considered to be extinct in the wild, and why this extremely vulnerable species may be the perfect focus species for student-aquarium collaborative conservation.

Best of RAW Winner

Utilizing the Beloved Seahorse to Promote Marine Aquaculture, Biology and Conservation

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Due to their unusual attributes and charismatic nature, seahorses are popular animals exhibited in public aquariums. By using seahorses as a model species, we can engage students in aquarium science and marine conservation efforts. This iconic fish has been feeling the strains of several environmental and human effects on its population such as loss of habitat, and coastal run off. With recent advancements and new technologies in the aquaculture world, captive breeding and been applied as a key tool for conservation. Current projects include incorporating light-emitting diodes (LED) to evaluate carious light wavelength intensities, retinal mapping, and population genetics. With these results, we hope to enhance the knowledge of rearing seahorses, gain a better understanding of their wild and captive populations, and educate others about career opportunities in marine conservation.

Wednesday, March 23
Session 2: Animal Transport

Accident and Rescue Management of Large Marine Animal Transports

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On June 10, 2015, the Dynasty Marine Transport Team loaded 4 adult sandbar sharks into our 18-wheel reefer configuration-temperature controlled transport trailer at approximately 9:00am and left our Marathon facility enroute to the New York Aquarium. Slightly north of Orlando approximately 6 hours later, the tractor blew a front tire. The tire wrapped around the truck's steering arm as it disintegrated off the wheel, thus freezing the steering system and eliminating any control of the vehicles trajectory. This caused the rig to skid off the turnpike and onto the grassy-soft shoulder of the highway. The momentum of the truck was finally halted

abruptly as it slammed into a large tree. Fortunately, none of the transport team was injured in the crash.

During the collision with the tree, both transport tanks broke free of their internal restraints and penetrated the front of the aluminum trailer. Further both transport tanks cracked and one shark was completely ejected out of the front of the trailer. A second shark was partially ejected. The transport team was able to quickly lift the shark that was partially ejected back into one of the damaged transport tanks. Since both the truck and trailer were a total loss and inoperable, the transport team immediately faced a life threatening emergency for the animals. As per our ever-present contingency planning, we called colleagues in the Orlando area to see if anyone could assist us in the rescue of the sharks. SeaWorld Orlando was able to quickly prepare and execute an emergency evacuation plan that included transport trucks, transport tanks, seawater, spare oxygen, staff and temporary holding space for the sharks – all prepared with great skill and with no advance notice. The execution of that plan and the eventual success of the rescue will be discussed in detail along with challenges associated with the brief, yet intense media scrutiny.

Aquarium on Wheels at the Aquarium of the Pacific

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The Aquarium of the Pacific in Long Beach, California, has operated the Aquarium on Wheels program since 2002. On average, the Aquarium on Wheels truck drives 10,000 miles each year while visiting 40,000 guests in and around the Long Beach area. This mobile tidepool contains all the essential infrastructure and life support systems to provide a permanent home for a variety of local species.

From October 2015 to January 2016, the Aquarium on Wheels vehicle has undergone an extensive renovation including redesigned, modified, and updated life support systems. Over the past 14 years, there have been several problems that our staff has addressed. Some problems include vibrations from driving affecting the life support systems, high levels of *Enterococcus* bacteria, water loss when driving, and bulky life support equipment. The new truck is specifically designed to address the common problems faced by the staff operating the truck when offsite. The new system design incorporates updated life support technology and provides a more user-friendly system while maintaining the same water quality and animal health standards as all other exhibits at the Aquarium of the Pacific.

Collection andTransport of *Sphyraena viridensis*

João Correia
Flying Sharks

Thursday, March 24
Session 1: Natural Disaster Preparedness Panel Discussion

The Regional Aquatic Workshop consistently draws aquarium professionals with a wide range of expertise and experience. Every aquarium facility strives to prepare for the most probable natural disasters in their geographic area. It is important to consistently share emergency preparedness procedures with younger aquarists, new managers, and also share new situations from veterans in the field.

This in mind, a panel of aquarium representatives will provide a framework for facilitated discussion of Disaster planning and response. Each of the six panel members will present a five minute introduction of their experiences in natural disasters: what worked, what did not, and how we've changed. This will lead into an open discussion with the audience, addressing concerns and ideas relevant to their facilities.

Panel Members: Jim Prappas, Director of Biology, Houston Aquarium, Inc.; Dr. Kenneth Yates, PhD, Life Sciences Manager, Landry's Downtown Aquarium – Denver; Greg Whittaker, Animal Husbandry Manager - Moody Gardens, Rainforest & Aquarium at Moody Gardens, Inc.; David DeNardo, General Curator/Dir. of Animal Operations, Wildlife Conservation Society-New York Aquarium; Kenny Alexopoulos, Director of Husbandry, Oklahoma Aquarium; James Arnold, Sr. Curator, Audubon Aquarium of the Americas.

Thursday, March 24
Session 2: LSS and Quarantine

Emerging Techniques for Quantifying Environmental Quality

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As an industry we are accustomed to measuring 'standard' environmental parameters for the animals in our care, such as nutrients, temperature, pH, etc. However, there are a number of other more esoteric, yet cheap and easy to measure, parameters that can give great insight into the quality of the environment. Examples include: atmospheric CO2, pH differential, and sand grain angularity. In addition, the development of mechanisms to quantify otherwise descriptive data can provide a valuable means to reveal trends and target trouble-shooting activities within living systems. Two examples include the development of a disease condition scale and the examination of relative biological loads within a dynamic system. Critical to the successful implementation of these techniques is a robust mechanism to feed collected information back into the operation of the living systems.

Water Quality Optimization and Resource Preservation Through Better Life Support System Design

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Equipment instrumentation indicates life support system *function*, but it is water quality parameters that indicate treatment *efficacy* and resource allocation that indicates treatment *efficiency*. Equipment parameters—pressure, flow and water level—can be operating within "design specifications", yet water quality parameters (e.g., nutrients, alkalinity, pH) can be unsuitable for aquatic life and excess resources (e.g., water, energy, labor) must be expended to normalize the environment. Two common water quality challenges have their root causes tied to poor life support system design and operation. Low pH and alkalinity occur when there is no means to export CO2 or when gas exchangers are poorly designed. High nitrates, a common reason for high volume water exchanges, results from insufficient assimilation, exportation and/or denitrification. Retrofitting suitably sized and designed gas management towers and denitrification systems can significantly reduce water, labor and energy consumption, while significantly improving conditions for aquatic life.

Praziquantel Dosing and Breakdown in Quarantine

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Praziquantel is an important medication used in Aquaria to treat monogenean flatworm parasite outbreaks. Due to the expense and difficulty of measuring Praziquantel in aquarium waters there is much that is not known about its persistence and breakdown when administered directly into aquarium systems. A quarantine system at the Aquarium of the Pacific was monitored under different circumstances to determine the most efficient dosing method and a way to delay the breakdown of Praziquantel. The breakdown products are evaluated using spectroscopy. Quarantine treatment options are reviewed and discussed.

Inorganic Phosphates in Reef Aquariums

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Inorganic phosphates in reef aquariums can often exceed values found on natural reefs, and are sometimes associated with decreased coral growth and coloration, and increased nuisance algae and cyanobacterial growth. Phosphate concentrations greater than 0.03 mg/L can be an indicator of poor water quality. Here we present information on various methods to reduce phosphates in a twenty-one-year-old 6,400-gallon reef aquarium with limited prior history of phosphate exportation and simplified life support. Specifically, we designed and built a side-stream filtration method that uses a commercial pool phosphate remover, Lanthanum Chloride,

to remove bulk phosphates from the system. Phosphate was reduced from 1.5 mg/L to 0.5 mg/L over a 6-month period. An analysis of our water quality data over this time period reveals insights into how to best use Lanthanum Chloride on a mature reef system that lacks cutting edge life support elements. We also examine methods to avoid potential pitfalls associated with Lanthanum Chloride use in aquaria.

Microbes and the Aquarium: Who's eating the Prazi?

Larry C. Boles, Amber Thomas, Matt Dawson, Helen Ellis,
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The effect of added chemicals on biological filters has been studied for many years and is the most important consideration when using therapeutic agents in recirculating aquarium systems. Drugs can also be utilized by microbes as a food source and may impact treatment procedures. Praziquantel (PZQ) is a drug commonly utilized to treat parasitic infections in humans and other animals. In the aquariums, PZQ is administered in a variety of ways ranging from short-term baths to long-term immersions for the treatment of ectoparasites on fish and elasmobranchs. In order to fully treat an infection, the aquarist may have to maintain therapeutic levels of PZQ over a period of many days. It has long been assumed that once administered, PZQ is stable in a marine environment and must be removed by filtration. However, no controlled experiments have been published to validate that claim.

In this study, recirculating marine aquariums (three containing 12 French grunts, *Haemulon flavolineatum*, and three with no fish) were treated with PZQ at 2 ppm. After one round of treatment, the PZQ was no longer detectable in any system after 8 days. There were no significant differences in concentration over time between systems with or without fish, suggesting that the removal of PZQ from the environment was not due to the presence of fish. The subsequent two PZQ treatments yielded even faster breakdown (non-detectable after 2 days) with slight variations between systems. The variability in breakdown rate suggests that mechanical filtration is likely not causing the breakdown. In sterilized system water, PZQ concentration remained unchanged over 15 days, suggesting that breakdown is not naturally-occurring in salt water, but rather may be the result of bacterial degradation. These results should be taken into consideration when providing PZQ treatments to marine animals to ensure maximum drug administration.

Successful Treatment of *Eimeria southwelli* in a Cownose Ray (*Rhinoptera bonasus*) using Oral Copper Wire Particles.

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Cownose rays (*Rhinoptera bonasus*) are medium sized elasmobranchs commonly kept in aquaria throughout North America. *Eimeria southwelli*, an apicomplexan parasite, is a known

organism associated with mortality in cownose rays.[1] Clinical signs of eimeria infection include discoloration, emaciation and death.[1] An adult female cownose ray housed in captivity long term, but initially wild caught was maintained in a large approximately 510,300 L multi-species exhibit. At initial clinical presentation, this ray was observed to be severely emaciated (weight 1.36 kg) and discolored. The animal was manually captured and medically evaluated. A coelomic cavity saline flush and sampling with cytological concentration was performed and *E. southwelli* oocysts were observed on microscopic evaluation of the fluid. The animal was treated with a one-time dose of copper wire particles (50 mg, Copasure®, Animax Ltd, Shepards Grove West Stanton, Bury St. Edmunds, Suffolk, IP31 2AR, England) at a dosage of 36.7 mg/kg. The copper wire particles were placed in an empty dosing gel capsule and fed orally with frozen-thawed diet items. Weights were obtained weekly, and coelomic cavity saline flush and sampling were performed at 6 and 10 weeks post copper wire treatment to observe for *E. southwelli* oocysts, both were negative for visible oocysts. At week 10 the animal had returned to a normal body condition, normal activity level, and was over 2 kg and determined to be clinically normal and returned to the main exhibit, where it remains. Coelomic saline sampling is one of the most common ways to diagnose *E. southwelli* in cownose rays.[1] In the case of this particular cownose ray, copper wire particles given orally appeared to be a safe and effective treatment option for an *E. southwelli* infection. Future studies evaluating the pharmacokinetic and dynamic effects and safety of this treatment in cownose rays is indicated.

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Topical Applications of Misoprostol and Phenyton Gel for Treatment of Dermal Ulcerations in Teleosts

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Misoprostol, a synthetic prostaglandin E_1 analog, is used to decrease gastric secretion and increase uterine contraction frequency in both veterinary and human medicine. Phenytoin is used as an anticonvulsant and antidysrhythmic, and is commonly incorporated into euthanasia solutions in veterinary medicine. These drugs have been compounded together and used topically to treat ulcers in human medicine. Mechanisms of action include stimulation of fibroblasts, increased collagen deposition, glucorticoid antagonism and antimicrobial activity. Misoprostol/Phenytoin gel (MP) was used for treatment of dermal ulcerations due to conspecific aggression and/or environmental trauma in multiple teleost cases at the Audubon Aquarium of the Americas and its effectiveness was evaluated.

A community compounding pharmacist prepared the two drugs into a fine powder, consisting of 0.0024% Misoprostol and 2% Phenytoin. During treatment, the affected teleost was manually restrained or sedated with tricaine methanesulfonate, and temporarily removed from the water. Lesion sites were cleaned with dilute chlorohexidine and rinsed with saline. A thin layer of MP was applied to the ulcerated area. Once in contact with moist surfaces, the powder quickly transformed into an opaque, sticky gel-like substance. Additional saline or enclosure water was applied to further transform the powder to gel if needed. Once a thin layer was established, the patient was returned to the normal enclosure. Depending on the lesion site and

application thickness, the compounded gel typically remained in place for 3-5 days, and re-application was performed as needed. The gel acts as a protective layer, potentially decreasing osmotic stressors to the patient while promoting granulation tissue and healing. This compound has been used in multiple marine and freshwater species. No adverse reactions have been noted due to application of the drugs in any patient. Misoprostol/Phenytoin gel is easily applied, durable in the aquatic environment, and subjectively decreases healing time in ulcerated tissue of teleosts.

Best of RAW Winner

Experimental Quarantine Procedures for *Heteractis magnifica* and *Stichodactyla haddoni*

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There is very little literature about common disease processes within anemones and even less about potential therapeutic modalities to treat and acclimate these animals in captivity. We attempt to classify and construe a quarantine protocol based on past experiences, anecdotal information, and histopathology reports.

Over the span of a year, two anemone species, *Heteractis magnifica* and *Stichodactyla haddoni*, were obtained from various marine animal suppliers. Animals were isolated into a 37.8 L glass enclosure which was placed in a 378 L fiberglass tray filled with water to help regulate temperature. The animals were given PVC pipe connections to attach to. A small powerhead separated by egg crate was also put into the enclosure to provide flow over the animal. Animals that died during the process were sent off for histopathology. Based off this information a quarantine protocol involving high flow, good water quality, and antimicrobial immersion bath therapies were developed. Antimicrobial therapy included immersive baths of Ciprofloxacin at 6.6 mg/L for 24 hours for 7 days with 50-100% daily water changes based on water quality. This was followed by an additional 3 days of observation with 50-100% daily water changes. The anemones were not fed during this time. Pictures of the anemones were taken and given daily scores on a scale of 1-10 based on observed health. A score of 1 being perfectly healthy and a score of 10 being nearly deceased. After 10 days of treatment, anemones with scores ≤ 2 were placed in mature holding systems and fed 3-4 times per week. Anemones were considered clear and healthy after one month of consistent feeding and inflation. While the use of Ciprofloxacin did appear to have a positive effect on the acclimation of anemones to captivity, the authors of this paper cannot stress enough the importance of responsible suppliers. Though all anemones received showed signs of shipping stress and irregular inflation and deflation, not surprisingly the highest survival rates were associated with animals that arrived inflated. Histopathology of submitted samples revealed ciliate organisms, amoeba, bacilli bacteria, and cestodes. This study attempts to describe potential pathogens affecting anemones and possible therapeutic modalities for acclimation and quarantine protocols.

Keeping you in the LOOP – a Unique Approach in treating Zebra Shark (*Stegostoma fasciatum*) Caudal Injury using Targeted Pulsed Electromagnetic Field Therapy and the ASSISI LOOP

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The Minnesota Zoo has a male zebra shark (*Stegostoma fasciatum*) which presented with a larger wound on the top of its caudal fin, a result of a small abrasion being picked on and worsened by numerous butterfly fish. After being placed in a holding pool, healing was very slow despite dosing with Baytril and using other conventional treatments. Our veterinarian staff and team of aquarists decided to try a unique medical device, called the ASSISI LOOP. The LOOP uses Targeted Pulsed Electromagnetic Field Therapy (TPEMF) to aid in soft tissue trauma and wound care. The results were very encouraging, as blood flow to the wound was immediately noticeable during treatment, and overall recovery time was reduced. Using the LOOP was quite simple. Once the shark was placed under tonic immobility, the injured portion of the tail was placed inside the loop to undergo the TPEMF treatment for a total of 15 minutes, once a week. Future use of the LOOP can be very practical in the aquarium industry. It is compact, safe, and can reduce or eliminate the use of pharmaceuticals, while expediting healing time on a variety of elasmobranch injuries.

Friday, March 25

Session 1: Exhibit Design and Lighting

Moon Jellies a Touching Experience

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The Aquarium of the Pacific has operated a popular Moon jelly touch exhibit for the past 5 years. The design was a retrofit that remained intact through multiple themed exhibits within our changing gallery. The time has come to pack it up for a new theme, but we've decided to design a new and improved permanent Moon jelly touch system that will open later this year. In this presentation I'll share the logistics involved with operating this touch system, what issues we hope to overcome with the new design, and our guest's response.

From Artificial to Live: The Design, Construction, and Maintenance of a Large Scale Live Coral Exhibit

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The Aquarium of the Pacific recently undertook the challenge of converting an 11,000 gallon (42,000 Liter) fish and artificial reef exhibit into a live coral reef tank. Substantial

changes had to be made by the Husbandry, Life Support, and Facilities departments in order to make this possible. Light fixtures were replaced with a combination of metal halides and LED's. The plenum was removed, and water flow was redesigned. New reactors were added to help with the water quality needed to maintain a living reef. Thousands of pounds of rock were added to make shelves for the coral to be placed. After five months of work, the exhibit was ready to be stocked with live coral. The Aquarium of the Pacific staff learned many important lessons from this process, and ended up with a thriving living coral reef exhibit.

The Science of Light

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"The Science of Light" A comprehensive look into the fascinating world of light. The talk would include an interactive (Yes the attendees participate) talk/demonstration about Spectroradiometer, Quantum flux (PAR), and other light meters commonly used to measure and categorize light for scientific use. During the talk I demonstrate a Spectroradiometer and a Quantum flux meter and do comparative measurements real time. The spectral graph is actually displayed live through the video projector. The talk would cover Kelvin, PAR, Spectral power distribution, Lumens, Lux, C.R.I, wattage and other commonly used/misused lighting terms. Attendees will learn about their relevance in evaluating light sources for exhibit lighting.

In the talk I cover color temperature and how Kelvin is often misused to categorize lamps. I discuss PAR and explain the common misconceptions when using PAR to evaluate light sources. I also prove scientifically why you should reconsider the role a PAR meter plays when evaluating light sources for photosynthetic organisms. I explain Spectral power distribution and what those colorful spectral charts actually mean. I also explain how to use the charts to compare light sources of different wattages/intensities. I discuss/compare human eye response and absorption wavelengths/efficiencies of photosynthetic organisms. Lastly, I discuss radiometric (wattage) vs. photometric (optical output) comparison

LEDs - Not Another Talk. More Information Leads to the Death of Metal Halides

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As more and more public aquariums are moving from metal halides to LEDs the number of manufactures trying to break the 1000 watt comparable units has never been higher. There has been so much advancement in the LEDs for coral husbandry in the past 4 years. The focus of major manufacturers has changed to larger and brighter units to take over the public aquarium markets. Our facility has been solely lit by LED for over a year now and the data is in. From corals to freshwater planted exhibits.

This year will focus on can LEDs sustain larger colonies of corals and penetrate to the inner branches? Are the LEDs holding up? Are the LEDs still producing all the photons that they

originally did? Are the LEDs keeping the color spectrum they originally were? Are they worth the investment? Can you really save money by switching to LEDs?

This year will have pictures, videos, and more information to build on what information has been presented in the past. There have been some good times and some not so good times with testing these LED units over larger public aquarium reefs of all sizes and depths.

We are working with many different manufactures to build units that can penetrate to the depths comparable to the 1000-watt metal halides while also keeping the spectrum we want. At the same time the affordability or comparability to running metal halides. With giving them the data they need and the specs we need this part of coral husbandry has made leaps and bounds. The LEDs have also been moving into the non-reef sector of lighting aquariums due to the cost savings and many other advantages over conventional lighting options.

Friday, March 25

Session 2: Training and Enrichment

Improving the Lives of 'Touch Tank' Elasmobranchs Through Enrichment and Training

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The goal of a behavioral enrichment program for aquatic species is the same as that of a terrestrial enrichment program; i.e., to elicit natural, species-appropriate behaviors while reducing stereotypy.

A 'touch-tank' is a type of zoo and aquarium exhibit whose purpose is to provide the visitor with a closer, more intimate experience with an animal species, however, it does not often reflect the natural environment nor encourage species-appropriate behaviors. Furthermore, animal behavior in 'touch-tank' exhibits can be influenced by guest interactions; e.g., hand feeding near the surface of the water. The behavioral observations within a population of 31 *Rhinoptera bonasus* led to the development of a new enrichment and training program at the Phoenix Zoo's Stingray Bay. The program attempts to maintain a balance between stimulating natural behaviors and providing a more rewarding visitor experience that enhances the connection between the guests and the animals. Interactions between the animals and their new enrichment were recorded, evaluated, and an ethogram was developed based on consistent behaviors seen within the population.

Getting to know Gerry: Operant Conditioning of a Giant Sea Bass

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Operant conditioning is a common form of training utilized by many working in the animal husbandry industry. Through the use of positive and negative reinforcement, one is able to achieve a desired behavior. Documented training has occurred amongst a variety of animals. From determining the intelligence of Crows (Powell, 1972), to conditioning parrots to receive medical treatments (Daugette, 2012), to even training dolphins to fight off sharks (Irvine, 1973). However, there is little documented success with teleost fish in the aquatic husbandry field. In this study, positive reinforcement is used to target train a Giant Sea Bass (*Stereolepis gigas*) in order to easily administer necessary medical treatments during outbreaks of a common eye parasite. Training was performed, in an isolated holding tank, three times a week for approximately 10 months to achieve the desired behavior. Training was then successfully continued after moving the animal back into the Kelp Tank Exhibit. This study gives evidence towards the intelligence of such a species and the ability for aquarists to perform less stressful medical applications with animals of the teleost class.

A BRIEF GUIDE TO AUTHORS

Updated 2017

This guide is intended for those not accustomed to using a "Guide to Authors", as provided by more formal periodicals. Historically only about 5% of *D&C* authors get this correct ☺. Please help me out folks!

As always, typical Drum & Croaker articles are not peer reviewed and content will not be edited, other than to correct obvious errors, clarify translations, modify incorrect or cumbersome formatting, or delete superfluous material. Other types of contributions (announcements, etc.) may be edited to meet space limitations.

The approximate deadline for submissions is December 15th. As has always been the case, materials in *Drum and Croaker* may be reproduced unless otherwise specified. Please credit *Drum and Croaker* and the contributor. I expect and assume that all submissions to D&C (papers, photographs, etc.) have been authorized by all original authors or co-authors, do not infringe on any copyright or prior publication agreements, and have successfully completed any internal review process required by your institution.

Submit articles via email as a Microsoft Word document (or a file that can be opened in Word). **My E-mail address is petemohan@aol.com.**

All Articles Must Adhere To The Following Basic Format:

- Use Times New Roman 12 point font throughout (except figure and table legends as noted below).
- A4 users please reformat to 8 ½ x 11 inch documents.
- Keep the resolution of photographs LOW. High resolution photos make the PDF file huge and are compressed anyway.
- **Format the title section with the line spacing set on 1.5 lines (not another method) and using centered, boldface font. Only the title should be CAPITALIZED (except italicized *Scientific namesii*)**.
- Double-space after your "institution name" to begin the body of your text. It should look like this:

USE OF DUCT TAPE IN THE HUSBANDRY OF *Genus species* AT FISHLAND

Jill Fishhead, Senior Aquarist jfishhead@fishstinking.com

Fishland of South Dakota, 1 Stinking Desert Highway, Badlands, SD, USA

Continued….

Text Format

Headings and text should look like this heading and paragraph. Use single spacing with 1" (2.54 cm) margins on ALL sides. Please indent 0.5 inch (1.3 cm) at the beginning of each paragraph and leave a space between paragraphs. Justify the text (see toolbar options and note how pretty the right margin of this paragraph lines up!). Section headings should be in bold (as above) at the left margin.

Figure Legends

Please use the following format:

> Figure 1. Legends should appear under the photo or graph in this format in 10 point font, aligned with the sides of the image or figure (center or justify). Photographs should be pasted into the document in the proper location by the author. All photos MUST be formatted as low resolution files, no 'larger' than approximately 300 – 500 KB. I may reduce the size (appearance on the page) of figures and photographs to save space. Photos, tables, and figures not referred to in the text may be omitted for the same reason.

Table Legends

Table legends go above the table. Otherwise, formatting is as above for figures.

Other Things I Whine About

- Please don't use Paragraph formatting to add space above or below lines. I have to remove all of these. Start with a single spaced Word template.
- Use the "enter" key for all spaces ("carriage return" for those who remember typewriters with a slidey thing on top).
- If you submit a table, put the data IN an actual table. Don't use the space bar or tabs to "line up stuff." This formatting can be lost if I have to change margins.
- Use the "tab" key to set your 0.5" indent at the start of each paragraph. It's likely your default. Don't use the space bar.
- Use bullets or numbers to make lists. It is easier to reformat these later if needed.

Short Contributions ("Ichthyological Notes")

These include any articles, observations, or points of interest that are about a page or less in length. A brief bold faced and capitalized title should be centered, the body text should be formatted as above, and **author and affiliation should be placed at the end of the piece** with the left end of each bolded line right of the center of the page. Reformatting that must be done by the editor may reduce a shorter "main" article to a note, or may bump a note up to main article status.

Reviews, abstracts, translations (with proper permissions) and bibliographies are welcome. Humor, apocrypha, and serious technical articles are equally appreciated.